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THE
DESIGN AND ANALYSIS
OF
FACTORIAL EXPERIMENTS

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The Design and Analysis of Factorial Experiments

I. INTRODUCTION.

Factorial experiments are experiments which include all combinations of several different sets of treatments or "factors." Information is thus simultaneously obtained on the responses to the different factors, and also on the effects of changes in the level of each factor on the responses to the others.

This Technical Communication has not been written with the object of convincing experimenters of the need for employing factorial designs, but rather for those who, while fully conscious of the advantages of such designs, find difficulty in laying them out and in analysing the results. It is, in fact, an attempt to give a comprehensive survey of the simpler types of design at present available, and a description of the appropriate methods of analysis. The reader who has not done so is advised first to read Prof. R. A. Fisher's *Design of Experiments*, where he will find a full account of the logical basis of the whole technique of modern experimental design.

1a. Principles underlying factorial design.

The points at issue may be made clear by the consideration of an example. Suppose it is desired to introduce a new crop into a country, and that nothing is known of the most suitable varieties, type and quantity of manuring, the best cultivations, etc. The classical procedure would be to set up separate experiments to determine the best varieties, others to investigate the manurial requirements, others (if indeed any were undertaken on this point) to determine the most suitable methods of cultivation, rates of sowing, etc. Unfortunately, however, we cannot conduct manurial experiments without choosing some variety on which to conduct them, nor can we conduct varietal trials without deciding on some level of manuring, a rate of sowing, width between rows, cultivations, etc. Now it may happen that the effects of fertilizers on the different varieties are materially different, or that varieties that are good yielders at wide spacings, owing to a rank habit of growth, are much inferior in yield (or other qualities) when sown at close spacings. Thus conclusions that have been laboriously reached on the correct level of manuring for one variety may be inapplicable to the variety finally chosen, and that variety may itself be incorrectly chosen through not realizing the possibilities of increasing the yield of other varieties by changes in cultural practices.

Of course none of these misfortunes may occur. The varietal differences may be substantially the same for all levels of manuring and all cultural practices, and responses to fertilizers may be unchanged by change in cultural practices. Indeed such experimental programmes would be completely futile were this not usually the case. But even where it does happen that no disturbances of this kind exist, such methods are exceedingly inefficient compared with factorial experiments, for the reason that in factorial experiments all the plots are used

many times over in making estimates of the effects of the different factors. Thus, for example, with four factors, each at two levels, there are 16 treatment combinations. With 80 plots five replications of each combination are therefore possible. The estimate of the effect of any one factor, if this effect is unchanged for variations of the other factors, is obtained from the comparison of the mean of the 40 plots receiving the higher level of this factor with the mean of the other 40. If four separate experiments are undertaken, one on each factor; then each experiment will contain 20 plots only, and the estimate of the effect of each factor will be obtained from the comparison of two means of 10 plots each. The precision will therefore be one quarter that of the factorial experiment, provided the standard error per plot is the same in both cases. Even if these four experiments are combined and one set of plots is used for the "controls," i.e. the plots receiving the standard level of each factor, there will only (with 16 "controls") be 16 plots for each factor, so that the precision will be $\frac{2}{5}$ that of the factorial design.

If the effects of some or all of the factors vary with changes in the other factors, the factors are said to *interact*, and the estimates obtained as above from a factorial experiment will be the *average* of the effects of each factor in conjunction with the different levels of the other factors. At the same time estimates of the actual amount of the variation may be obtained by taking the differences of the effects of one factor at the different levels of the other factors. In such circumstances the results of a set of experiments containing single factors only will be misleading to an extent depending on the degree of variation in the effects.

1b. Criticisms of factorial design.

It is sometimes objected that what is really required is not the *average* effect of a factor, but rather the effect of this factor in conjunction with some particular combination of the remaining factors, and that factorial experiments provide an estimate of this having only low precision. Actually it rarely happens that agricultural practices are in fact standardized in the way contemplated by the critics, but even where this is the case the objection, as we have seen, carries no weight unless the variation in the effects is substantial, and even then the loss in precision is small if the levels of the remaining factors finally adopted are intermediate between the extremes included in the experiment. In any case unless we know beforehand the particular combination of the other factors that will be used (in which case it will be a waste of time experimenting on them at all) we are forced to survey the whole field, and the experimenter who confines himself to experiments on single factors, making a guess at the final levels of the other factors, is merely emulating the tactics of an ostrich.

An objection of a similar type is that such and such a combination of factors "would never be used in practice." Thus in fertilizer trials it may be maintained that the application of phosphate without potash or nitrogen to a certain crop is ridiculous. Such preconceived notions are usually based on entirely inadequate evidence, and are well worth experimental test, but as evidence accumulates the field of enquiry can sometimes profitably be narrowed. Thus if it is known

that the application of some nitrogenous manure is certainly required, but the optimal level is still in question, the lowest level of nitrogen need not be zero, but a minimal dressing. There is also no objection in randomized block experiments to including an additional set of plots (outside the main factorial scheme) receiving no nitrogen, both for demonstration purposes and as an assurance that conditions have not radically changed.

There is one further point which must be considered in assessing the advantages of designs of varying complexity. As the number of treatment combinations is increased the adequate elimination of fertility differences becomes more difficult. Consequently the standard error per plot tends to be higher in factorial designs than in simple experiments involving a few treatments only, with a resultant lowering of the relative efficiency of factorial designs. The whole matter has been discussed in (9)* where it was shown that the loss of efficiency with properly designed experiments may be expected on the average to be much less than the gain due to the use of factorial design, quite apart from the information on the interactions between the different factors, which can only be obtained from factorial designs. The loss of efficiency was found to be due mainly to the necessity of abandoning Latin-square arrangements, the discussion being written before it was realized that Latin-square designs could be utilized in some types of factorial experiments. This procedure, when it is possible, is likely to reduce the loss materially.

It is, perhaps, typical of the superficial character of most criticisms of factorial design, that in many of them the efficiency of a design (i.e. relative amount of information per unit of work expended, or per plot when the work expended is proportional to the number of plots), is confused with the accuracy of the final comparisons, which accuracy can always be increased by increasing the size of the experiment, and therefore the number of replications, or by decreasing the number of treatments included in the experiment.

The difficulties of the practical type that stand in the way of factorial design arise from the greater complexity both of the layout and the statistical analysis, and the larger number of plots that are required. How far these are of importance must be decided by the man in charge of the field operations. In this connection it should be remembered that any new technique is liable to present difficulties which fade away on closer acquaintance.

1c. Scope of the Communication.

In the present paper factorial designs with factors at two levels only are first discussed, since these are capable of specially simple treatment, and enable the structure of confounded arrangements to be more easily understood than do designs containing factors at three or more levels. There follows an account of designs with factors at three levels, with factors both at two and three levels, and with factors at two, four and eight levels. Finally, various special types of design, such as designs with split-plots, and their modifications, and designs for varietal trials involving a large number of varieties, are described.

*The numbers refer to the references at the end of the paper.

No attempt has been made to give recommendations as to the best procedure in the field, or to discuss such points as size and shape of plot, number of replications, etc., since these depend so much on type of crop and local conditions that no discussion in general terms would be profitable. It may be well to emphasize here, however, that the additional complexity of factorial designs (and to a lesser extent all random arrangements) carries with it the necessity for careful organization if mistakes are to be avoided. The preparation of clear and simple plans, and a convenient system of numbering the fertilizer mixtures, etc., that are to be applied, will lighten the work of the man in the field, who is usually operating under adverse conditions, is frequently in a hurry, and is sometimes not very certain of the points at issue. Whenever the remark is heard, for instance, that random arrangements lead to mistakes in the field from which systematic arrangements are immune, it can be confidently predicted that the preliminary organization is inadequate.

Id. New material.

For the benefit of the reader who is already familiar with the subject it may be well to indicate here what is new in this communication. Most important is the adaptation of confounding to Latin-square designs, so as to enable, for instance, a 2^5 experiment to be arranged in the form of an 8×8 Latin square (pp. 31-35, etc.). The analogous adaptation of split-plot designs is also of considerable importance (pp. 78-81). The parallel use of quasi-Latin squares (lattice squares) in varietal trials (described in full elsewhere) is also outlined (pp. 87-8).

No complete account of the designs involving some factors at two and some at three levels (pp. 57-64) has previously been published, though some of these designs have been in use at Rothamsted and elsewhere for some years. The account of designs containing factors at two levels only (pp. 23-26) is also more complete than any previously published. Lastly, the 3^4 design in blocks of 9 plots (pp. 47-8), a fairly obvious extension of the popular 3^3 design, should be noted.

On the computational side a new method of computing the treatment effects in experiments with factors at two levels only is given (p. 15), and attention has been paid generally to the best methods of carrying out the computations of the various designs.

Id. Notation, etc.

It is assumed that the reader is familiar with the methods of design and analysis appropriate to simple experiments in randomized blocks and Latin squares, and in particular that he is thoroughly conversant with the *analysis of variance* procedure applicable to experiments of this type. A selection of references on the subject is given at the end of the paper.

The notation followed is substantially that of Fisher's *Design of Experiments*, i.e. small letters are used to denote the treatments corresponding to the different factors, and capital letters the main effects and interactions. The symbol $[ab]$ has been introduced to indicate the sum of all the yields corresponding to the treatment combination ab , the symbol ab , when it indicates a number, being

used to represent the mean of these yields, or this mean expressed in standard units (cwt. per acre, etc.). (In *The Design of Experiments* (ab) is used to denote either the sum or the mean according to the experimental material.) By analogy $[A]$ and $[A.B]$ are taken to represent the algebraic sums of all the plot yields which go to make up the estimates of the main effect of a and the interaction of a and b , without any division, whereas A and $A.B$ indicate these estimates expressed in terms of the yield of a single plot (or in standard units, such as cwt. per acre), with the conventional factor $\frac{1}{2}$, $\frac{1}{4}$, etc. introduced, as defined on page 10. In the case of factors at more than two levels the symbol $[A]$ is taken to represent the whole set of totals, and A the whole set of means, corresponding to the various levels a_0, a_1, a_2, \dots of the factor a .

One other new symbol is introduced. This is the word *dev*, which is used to denote the deviations of a set of numbers from their mean. dev^2 is likewise used to denote the *sum* of the squares of these deviations. Thus

$$\text{dev}(a_1, a_2, \dots) \equiv \text{dev } a \equiv a_1 - \bar{a}, a_2 - \bar{a}, a_3 - \bar{a}, \dots$$

$$\begin{aligned} \text{dev}^2(a_1, a_2, \dots) &\equiv \text{dev}^2 a = S(a - \bar{a})^2 = a_1^2 + a_2^2 + \dots - n\bar{a}^2 \\ &= a_1^2 + a_2^2 + \dots - \frac{1}{n}(a_1 + a_2 + \dots)^2 \\ &= a_1^2 + a_2^2 + \dots - \bar{a}(a_1 + a_2 + \dots) \end{aligned}$$

In a similar manner $\text{dev } a, \text{dev } b$ might be used in covariance work to indicate the sum of the products of the deviations of two variables a and b . The occurrence of these quantities in statistical computation appears to be sufficiently frequent to justify the use of a special symbol, especially since they are only very clumsily representable by the current symbols when the a 's are themselves complicated algebraic expressions.

Algebraic formulæ have been avoided as far as possible, and where it has been necessary to introduce them particular attention has been paid to writing them in the form required by the computer and also in a form exhibiting their structure, so that they are easily remembered. Thus the quantity Q on page 58 has been so defined as to be analogous with the quantity $[B.C]$, but the formula for $3Q$ is given because $3Q$ will be computed in numerical work. The formula for $B.C$ on the same page is given in terms of both Q and $3Q$, the latter being the form required for computation, while the former exhibits the structure.

Free use has, however, been made of the algebraic notation of signs, brackets, etc., in setting out arithmetical calculations. Those who can understand this notation (as for example the expression for the sum of squares on page 37) should have no difficulty with the algebraic formulæ.

2. A SIMPLE FACTORIAL EXPERIMENT ON POTATOES.

The main features of factorial designs involving only two levels (often presence and absence) of each factor can best be illustrated by a simple example. We have chosen an experiment on the manuring of potatoes carried out at Wimblington in 1934.

Three factors, nitrogen, potash and dung, were included ; the 8 treatment combinations consisted of all combinations of :

$$\left\{ \begin{array}{c} \text{Sulphate of Ammonia (n)} \\ \text{None} \\ \text{0.45 cwt. N per acre} \end{array} \right\} \times \left\{ \begin{array}{c} \text{Sulphate of Potash (k)} \\ \text{None} \\ \text{1.12 cwt. K}_2\text{O per acre} \end{array} \right\} \times \left\{ \begin{array}{c} \text{Dung (d)} \\ \text{None} \\ \text{8 tons per acre} \end{array} \right\}$$

There were four replications in randomized blocks of 1/60 acre plots. The plan of the experiment and the yields of the individual plots are shown in Table 1.

TABLE 1. PLAN AND YIELDS IN LB.

Block I				Block II				Block Totals			
<i>nk</i> 291	<i>kd</i> 398	<i>d</i> 312	<i>nd</i> 373	<i>kd</i> 407	<i>d</i> 324	<i>k</i> 272	<i>nk</i> 306	I	2296		
(1) 101	<i>k</i> 265	<i>n</i> 106	<i>nk</i> 450	<i>n</i> 89	<i>nk</i> 449	<i>nd</i> 338	(1) 106	II	2291		
<i>d</i> 323	(1) 87	<i>nd</i> 324	<i>kd</i> 423	<i>nd</i> 361	<i>nk</i> 272	<i>n</i> 103	<i>d</i> 324	III	2369		
<i>nk</i> 334	<i>k</i> 279	<i>n</i> 128	<i>nk</i> 471	<i>k</i> 302	(1) 131	<i>nk</i> 437	<i>kd</i> 445	IV	2375		
Block III				Block IV				Total	9331		

2a. Yields of the different combinations of treatments.

The first step in the analysis of the results of a factorial experiment is to calculate the total yields of all the plots with each combination of treatments. The main features of the results are usually apparent from an inspection of these totals. An analysis of variance will, however, be necessary in order to make the formal tests of significance and assign standard errors to the various comparisons.

The yields of the individual treatment combinations in this experiment (converted to tons per acre) are given in Table 2.

TABLE 2. YIELDS OF THE DIFFERENT COMBINATIONS OF TREATMENTS
(TONS PER ACRE).

(1)	<i>n</i>	<i>k</i>	<i>nk</i>	<i>d</i>	<i>nd</i>	<i>kd</i>	<i>nk</i>	Mean
2.84	2.85	7.49	8.06	8.59	9.35	11.20	12.10	7.81

Treatments are indicated by small letters, and the symbol (1) is used to indicate absence of all fertilizer.

2b. Main effects.

Consider first the effect of dung. There are four relevant comparisons in Table 2.

$$\begin{array}{l} \text{Response to dung} \left\{ \begin{array}{l} n \text{ and } k \text{ absent} = d - (1) = 5.75 \\ n \text{ absent, } k \text{ present} = kd - k = 3.71 \\ n \text{ present, } k \text{ absent} = nd - n = 6.50 \\ n \text{ and } k \text{ present} = nkd - nk = 4.04 \end{array} \right. \\ \text{Mean response} = D = 5.00 \end{array}$$

These large apparent responses are sufficiently consistent to indicate that they are unlikely to be due to experimental error.

The mean response, 5.00, will be called the *main effect* of dung, and will be denoted by the capital letter D .

In a similar manner we have

$$\begin{array}{l} \text{Response to potash} \left\{ \begin{array}{l} n \text{ and } d \text{ absent} = 4.65 \\ n \text{ absent, } d \text{ present} = 2.61 \\ n \text{ present, } d \text{ absent} = 5.21 \\ n \text{ and } d \text{ present} = 2.75 \end{array} \right. \\ \text{Mean response} = K = 3.80 \end{array}$$

$$\begin{array}{l} \text{Response to nitrogen} \left\{ \begin{array}{l} k \text{ and } d \text{ absent} = 0.01 \\ k \text{ absent, } d \text{ present} = 0.76 \\ k \text{ present, } d \text{ absent} = 0.57 \\ k \text{ and } d \text{ present} = 0.90 \end{array} \right. \\ \text{Mean response} = N = 0.56 \end{array}$$

There is, therefore, also evidence of a substantial response to potash, and possibly a small response to nitrogen.

2c. Interactions.

Examining the individual responses further, we see that the two responses to dung with potash absent are both substantially larger than the responses with potash present. Equally the responses to potash are substantially larger in the absence than in the presence of dung. The presence or absence of nitrogen, however, makes little difference in either case.

The numerical differences in response to dung in the presence and absence of potash are as follows:

$$\begin{array}{l} \text{Difference in response to } d \left\{ \begin{array}{l} n \text{ absent} \dots \dots - 2.04 \\ \text{in presence and absence of } k. \quad n \text{ present} \dots \dots - 2.46 \end{array} \right. \\ \text{Mean} \dots \dots - 2.25 \end{array}$$

For reasons that will be apparent in a moment it is convenient to take *one half* this mean difference, namely - 1.12. This is defined as the interaction between the two factors* d and k , and may be written $D \times K$, $D.K$ or DK .

*Also called the *first order* interaction. The mean interaction over all the other factors in the experiment is implied unless the contrary is stated.

A similar computation for differences in the responses to potash in the presence and absence of dung gives the identical results :

Difference in response to k in presence and absence of d .	n absent	- 2.04
	n present	- 2.46
Mean					- 2.25

A moment's consideration will show that this must be so. Thus the interaction between dung and potash is identical with the interaction between potash and dung.

An alternative method of setting out the main effects and interactions between two factors is by means of a two-way table. In this example, taking the mean of n and no n in each case, we have the values of Table 3.

TABLE 3. MEAN OF n AND NO n (TONS PER ACRE).

	No d	d	Mean
No k	2.84	8.97	5.90
k	7.78	11.65	9.72
Mean	5.31	10.31	7.81

The main effects are given by the differences between the pairs of marginal means, and the interaction is given by the difference of the means of the two diagonals, i.e. by $\frac{1}{2} (2.84 + 11.65 - 8.97 - 7.78)$.

In a similar manner, the difference between the values -2.46 and -2.04 gives an estimate in the change in the interaction between potash and dung in the presence and absence of nitrogen. *One quarter* of this difference, i.e. *one half* the difference of the interactions, is defined as the interaction between the three factors* and may be written $N \times K \times D$, $N.K.D$ or NKD .

2d. *Calculation of the main effects and interactions from the experimental yields.*

It will readily be seen from the above remarks that the main effects and interactions may all be obtained by subtracting the mean of 4 of the yield values of the individual treatment combinations from the mean of the other 4, or alternatively by taking the sum of 4 of these values less the sum of the other 4, and dividing the result by 4. The actual signs are given in Table 4.

TABLE 4. MAIN EFFECTS AND INTERACTIONS IN A THREE-FACTOR EXPERIMENT.

Effect	Combination of treatments.							
	(1)	n	k	nk	d	nd	kd	$nk d$
Total	+	+	+	+	+	+	+	+
N	-	+	-	+	-	+	-	+
K	-	-	+	+	-	-	+	+
$N.K$	+	-	-	+	+	-	-	+
D	-	-	-	-	+	+	+	+
$N.D$	+	-	+	-	-	+	-	+
$K.D$	+	+	-	-	-	-	+	+
$N.K.D$	-	+	+	-	+	-	-	+

*Also called the *second order* interaction.

These signs may be derived in various ways. The simplest is to write down the signs for the three main effects, and then to form the interactions between each pair of main effects by writing + for two +'s or two -'s, and - for a - and a +. A further application of this process gives the interaction between the three factors. If there are more than three factors the table may be extended by still further applications of the same rule.

The following formal expressions for the interactions are also worth noting :

$$\begin{aligned} N &= \frac{1}{4} (n-1) (k+1) (d+1), \\ N.K &= \frac{1}{4} (n-1) (k-1) (d+1), \\ N.K.D &= \frac{1}{4} (n-1) (k-1) (d-1). \end{aligned}$$

If these expressions are expanded by the ordinary rules of algebra the appropriate expressions for the main effects and interactions in terms of the treatment combinations will be obtained. With four factors the fraction will be $\frac{1}{8}$, with five $\frac{1}{16}$, etc., and with only two factors $\frac{1}{2}$.

If the above method of calculation be applied to our example the main effects and interactions will be found to have the values given in Table 5. Some of these have been obtained already.

TABLE 5. MAIN EFFECTS AND INTERACTIONS.

$N = +0.56$	$N.K = +0.18$	
$K = +3.80$	$N.D = +0.27$	$N.K.D = -0.10$
$D = +5.00$	$K.D = -1.12$	

A more mechanical method of obtaining these values is given in the next section.

These values clearly all have the same standard error, since they are each one quarter of the sums and differences of the yields of the eight treatment combinations. As we shall show presently, the estimate of this standard error (21 degrees of freedom) is ± 0.177 . Any value more than twice its standard error may be judged significant. Thus all three main effects and the interaction between potash and dung, the two factors producing the large effects, are significant.

This type of result is one commonly found in agricultural trials. Factors which produce large main effects may show evidence of interactions, but factors which produce small main effects usually show no significant interactions. A little consideration will show that this is what may be expected on general grounds. The interactions are in general likely to be small in comparison with the corresponding main effects.

2e. Interpretation of main effects and interactions.

It will be clear from what has already been written that the whole set of main effects and interactions, together with the mean yield, is equivalent to the yields of the individual treatment combinations.

The response to any factor or combination of factors in the presence or absence of any other factor or factors (the mean being taken over all factors not under consideration) can be written down very simply in terms of the main effects and interactions. The rules will be obvious from the study of Table 6.

TABLE 6. RESPONSES IN TERMS OF MAIN EFFECTS AND INTERACTIONS.

Response to :	Expression in terms of	
	treatment combinations	main effects and interactions
d (k absent)	$\frac{1}{2}\{nd - n + d - (1)\}$	$D - K.D$
d (k present)	$\frac{1}{2}\{nkd - nk + kd - k\}$	$D + K.D$
d and k together	$\frac{1}{2}\{nkd - n + kd - (1)\}$	$D + K$
d (n and k absent)	$d - (1)$	$D - N.D - K.D + N.K.D$
d and k (n absent)	$kd - (1)$	$D + K - N.D - N.K$
d , k and n	$nkd - (1)$	$D + K + N + N.K.D$

The interactions may thus be regarded as correcting terms which adjust the values of the main effects (which would be additive if the interactions were all zero). In this example the response to d where k is absent (mean of n and no n) is

$$D - K.D = + 5.00 + 1.12 = + 6.12$$

and where k is present is

$$D + K.D = + 5.00 - 1.12 = + 3.88$$

The response to both d and k (mean of n and no n) is

$$D + K = + 5.00 + 3.80 = + 8.80$$

These responses are those given by the differences of the values of Table 3.

It should be particularly noted that the interaction between d and k does not enter into the latter response. In the same way only the three-factor interaction enters into the expression for the simultaneous response to all three fertilizers :

$$D + K + N + N.K.D = + 0.56 + 3.80 + 5.00 - 0.10 = + 9.26$$

(This response can be obtained from Table 2.) If the interactions between the three factors were ignored, therefore, the estimate would be

$$D + K + N = + 9.36.$$

The yield of any treatment combination may also be obtained from the main effects and interactions, together with the mean yield, being equal to the mean yield and the sum of plus or minus *one half* of all the main effects and interactions. The signs are given by Table 4. Thus, for example :

$$kd = \text{mean} + \frac{1}{2}\{-N + K - N.K + D - N.D + K.D - N.K.D\}$$

It will be noted that in the order shown the table is symmetrical about the diagonal through the top right-hand corner, so that the expression for kd (equivalent to n absent) is obtained from that of N by replacing (1) by the mean, n by $\frac{1}{2}N$, etc., and changing signs if the sign of (1) is negative.

2f. General remarks.

The statement of the results in terms of main effects and interactions thus forms a convenient way of summarizing a factorial experiment, and concentrating attention on its main features. It should not be forgotten, however, that the expressions for the main effects and the interactions are really a matter of definition, the interactions being measures of the departure of the observed differences from the law implied in the definition of the main effects. Here the main effects are so defined as to imply an additive law between the effects

due to the three factors. This is statistically convenient, and in agriculture appears to provide a good representation of the type of effect frequently observed. But it should be clearly understood that the additive law has been provisionally imposed by the statistician and is not implicit in the data.

The present example has itself afforded an illustration of a simple type of departure from the additive law. Others more complex will occasionally arise, and the experimenter should then bear in mind that the formal presentation of the results in terms of main effects and interactions may not necessarily be the best course to pursue. Equally, however, he should avoid giving exaggerated emphasis to some statistically significant but isolated high order interaction which has no apparent physical meaning. If we are using the 1 in 20 level of significance one out of every twenty of the main effects and interactions will on the average be judged statistically significant even when the treatments produce no effects at all. Such anomalous results, therefore, together with non-significant effects, should be placed on record and judgment reserved until further information has accumulated.

Conversely, a verdict of non-significance does not imply that no effect exists. It merely implies that the observed apparent effect would arise more frequently than 1 in 20 (or 1 in 100) times by chance if there were no real effect. The application of exact tests of significance to all experimental results is a salutary habit which discourages the discussion of small apparent differences whose magnitude is very ill determined, but it should not be forgotten that the main object of most agricultural field trials is to *estimate* as accurately as possible effects of which the experimenter is normally quite prepared to admit the existence. A secondary requirement is the determination of the magnitude of the errors to which these estimates are subject, thus fixing limits between which the true value of the effect is likely to lie. Consequently tests of significance are replaced by estimates of standard errors and *fiducial probability*.

Thus, for example, it is reasonable to suppose that the application of nitrogenous fertilizer to a crop on a given area will always alter the yield of that crop, although the alteration may in certain cases be very small. Non-significant results must not be taken as implying that no effect exists in such experiments, though they can be taken as implying that the effect lies within certain limits. In conjunction with other results, also not in themselves significant, they may show quite clearly the existence of a small, but appreciable, effect. Similarly the practice of finding the average response to a fertilizer at stations where that response is significant is meaningless, for by making this selection of stations we automatically select a majority of stations at which the error in the estimated response is positive.

3. STATISTICAL ANALYSIS OF A $2 \times 2 \times 2$ EXPERIMENT.

The discussion in the last section was designed to illustrate the various aspects of the results of a simple factorial design. The routine analysis of such an experiment is, of course, much abbreviated, and in the present section we propose to give an outline of the various steps which should be followed in order to arrive at these results expeditiously and without unnecessary repetition of the various calculations.

1. *Yields of plots.* Set out the yields as in Table 1, rounding off, if necessary, to three significant figures. (See note 1, p. 91).

2. *Totals of individual treatment combinations and block totals.* These are shown in the yield column of Table 7 and in Table 1 respectively.

3. *Calculation of main effects and interactions in terms of the totals of the individual treatment combinations.* The main effects and interactions can be calculated from the totals of the individual treatment combinations by means of the table of signs in the last section. No division of the resultant totals need be carried out. These totals are shown in column (3) of Table 7, each being the sum of 16 plot yields less the sum of the other 16.

A more systematic and shorter method, which avoids the trouble of picking out the relevant treatment combinations (a process which is laborious when there are a large number of factors) is that shown in Table 7.

The yields must first be arranged in a *standard order* of the type shown, each factor being introduced in turn, and being followed by all combinations of itself and the factors previously introduced. Thus the last four combinations are formed by taking *d* in conjunction with the first four combinations.

Column (1) is then formed. The first four numbers are the sums of the four pairs of numbers in the yield column, and the last four numbers are the differences of these pairs, *the upper number being subtracted from the lower in each case*. Thus $2321 = 1118 + 1203$ and $+85 = -1118 + 1203$. Column (2) is formed in the same manner from column (1), and column (3) from column (2). Since there are three factors these three applications of the process complete the calculation. The total, and the main-effect and interaction totals, are obtained in column (3), each effect and interaction appearing opposite the corresponding small letters in the first column.

TABLE 7. CALCULATION OF TREATMENT EFFECTS.

Treatment	Yield	(1)	(2)	(3)	Effect
(1)	425	851	3172	9331	Total
<i>n</i>	426	2321	6159	+333**	<i>N</i>
<i>k</i>	1118	2679	+86	+2271**	<i>K</i>
<i>nk</i>	1203	3480	+247	+105	<i>N.K</i>
<i>d</i>	1283	+1	+1470	+2987**	<i>D</i>
<i>nd</i>	1396	+85	+801	+161	<i>N.D</i>
<i>kd</i>	1673	+113	+84	-669**	<i>K.D</i>
<i>nkd</i>	1807	+134	+21	-63	<i>N.K.D</i>
S.E.	±37.2			±105.4	
Conversion factor	60			60	
	2240×4			2240×16	
	=.00669643			=.00167411	

Significance levels (column 3): 5%: 219; 1%: 298

Asterisks denote significant results at 1% level.

There are no very simple checks on the intermediate stages of the calculation. Complete accuracy should therefore be aimed at, particular

attention being paid to signs.* The sum-of-squares check, described below, controls the whole calculation except for the signs of the last column, which should be independently checked. Interchanges in the yield column must be avoided by systematic computation. A useful partial check is provided by the sum, which is independently obtained from the block totals. This and the independent calculation of the interaction between all factors check all of the yield column and column (1), and one half of column (2).

A more elaborate example of the method, involving 5 factors, is shown in Table 22, where a systematic check for each column is introduced.

4. *Calculation of sums of squares for blocks, treatments, and error.* The ordinary methods of the analysis of variance are followed. These give the analysis of Table 8. It is advisable to record the correction for the mean as this is often required in subsequent calculations.

TABLE 8. ANALYSIS OF VARIANCE.

	D.F.	Sum of squares.	Mean square
Correction for mean ..		<u>2720861.3</u>	
Blocks	3	774.1	258.0
Treatments.. ..	7	458718.0	65531.1
Error	21	7287.6	347.0
		<u> </u>	
Total	31	466779.7	

5. *Partition of the treatment degrees of freedom and sum of squares.* The 7 degrees of freedom for treatments can be divided into 7 single degrees of freedom representing main effects, interactions between two factors, and the interaction between all three factors. The seven sums of squares may be calculated by squaring the quantities of Table 7, column (3). They are shown in Table 9.

TABLE 9. PARTITION OF TREATMENT SUM OF SQUARES.

	D.F.	Sum of squares
<i>N</i> ..	1	3465.3
<i>K</i> ..	1	161170.0
<i>N.K</i> ..	1	344.5
<i>D</i> ..	1	278817.8
<i>N.D</i> ..	1	810.0
<i>K.D</i> ..	1	13986.3
<i>N.K.D</i> ..	1	124.0
	<u> </u>	<u> </u>
Total	7	458717.9

Each square must be divided by 32, since it is the square of the total of ± 1 times the yields of each of 32 plots. (See note 2.) Thus

$$161170.0 = 2271^2/32$$

*The sum of two numbers of the same sign is the arithmetic sum and has itself this sign. The sum of two numbers of opposite signs is the arithmetic difference and has the sign of the larger number. To obtain the difference of two numbers change the sign of the number to be subtracted and take the sum as above. Examples will be found in Table 22.

These 7 degrees of freedom are *orthogonal* and therefore the sum of the 7 sums of squares is equal to the ordinary treatment sum of squares. (See note 3.) This provides the *check* of Table 7 mentioned above, and also checks the treatment sum of squares and the correction for the mean in Table 8.

Since the tests of significance can be performed by the *t* test (as described below) there is in practice no need to write down the separate sums of squares for each main effect and interaction, and Table 9 will consequently be omitted. All that is necessary is to sum the squares of column (3) of Table 7 (excluding the sum) on the machine, and divide the result by 32.

6. *Calculation of mean squares and tests of significance.* The separate components of the sum of squares for treatments can be tested for significance by means of the *z* test. Since in this case each corresponds to a single degree of freedom, however, it is simpler to use the *t* test, which is equivalent to the *z* test for $n_1 = 1$.

Since there are seven separate effects to be tested it is worth calculating the 5% and 1% points. For 21 degrees of freedom $t = 2.080$ for the 5% point and 2.831 for the 1% point. The estimate of the standard error of a main-effect or interaction total is $\sqrt{32 \times 347.0} = 105.4$. The 5% and 1% significance levels for the main-effect and interaction totals are therefore $105.4 \times 2.080 = 219.2$ and $105.4 \times 2.831 = 298.4$. Thus we see immediately that *N*, *K*, *D*, and *K.D* all attain the 1% level of significance, the remaining interactions not being significant.

7. *Conversion of yields and presentation of the results.* The yields should be converted to the customary agricultural units, and the results presented in the form most suitable for making clear the main features, and for combination with results of other experiments. Many alternative forms are possible, and the exact form will depend largely on circumstances. In general it is a good rule to present tables showing all main effects and interactions between two factors (and also any interactions between three or more factors which appear to be of interest) either directly or in the form of two-way tables with marginal totals. Various examples of the different types of presentation will be found in this communication.

The results of the present experiment have already been discussed in detail in the previous section. Both Tables 2 and 5 can be derived directly from the conversion of the appropriate columns of Table 7. Notice that the conversion factor for the effect totals of Table 7 is that applicable to the totals of 16 plots, although the effect totals each involve 32 plots, i.e. the difference of two sums of 16 plots.

The appropriate standard errors should be written in Table 7 and converted at the same time as the numbers to which they refer.

8. *Calculation of the yields of the treatment combinations from the main effects and interactions.* A procedure similar to that adopted for the calculation of the main effects and interactions from the yields of the individual treatment combinations is available for the reverse computation. This procedure is particularly useful in experiments involving a large number of factors when a

table giving the mean yields of the combinations of two or three factors averaged over the remaining factors is required. It can also be used to reconstruct the yields of the individual treatment combinations, when these latter are not available.

As an example we have derived Table 3 from Table 5 and the mean yield. The computations are shown in Table 10. Since only the two factors k and d are involved effects involving n will not enter into the calculation.

K , D , $K.D$ and *twice* the mean yield are arranged in a column, the order being the same as that adopted in Table 7, but beginning from the *bottom*. The computation process used in Table 7 is now applied. Only two applications are necessary as only two factors are involved. The last column is divided by 2 to give the required mean yields.

TABLE 10. CALCULATION OF YIELDS OF TREATMENT COMBINATIONS FROM MAIN EFFECTS AND INTERACTIONS.

				Effect	(1)	(2)	Yield		
$K.D$	- 1.12	+2.68	23.30	11.65	kd	} Mean over n and no n
K	+3.80	20.62	15.54	7.77	k	
D	+5.00	+4.92	17.94	8.97	d	
2 (Mean)	15.62	10.62	5.70	2.85	(1)	

As an exercise in the more extended application of this process the yields of Table 2 may be derived, using all the effects of Table 5.

4. CONFOUNDING.

Confounding is a device whereby the necessity of including every combination of the treatments of a factorial design in each block (or row and column in a Latin square) is avoided. This enables the block size to be kept small even when the number of treatment combinations is quite large.

In a confounded experiment the treatment combinations of each replication are divided into two or more groups (each group being assigned to a separate block) in such a way that the contrasts between the different groups represent high-order interactions, which as we have already seen are usually of less interest than the main effects and interactions between two factors only. Thus in any one replication the contrasts representing certain interactions are identified, or *confounded*, with the block differences, and in consequence in this replication most* of the information on these interactions is sacrificed. In so far as the reduction of block size has been effective in reducing the error variance the precision of all the remaining comparisons is increased. Moreover by confounding different interactions in different replications, i.e. by *partial confounding*, some information may be retained on all interactions—indeed if the gain in precision resulting from the confounding is sufficiently great even the partially confounded interactions may be more accurately determined than would be the case if the experiment were not confounded.

*A small amount arising from block comparisons remains, but is not in practice utilizable (see next page).

4a. Example to illustrate confounding.

A simple and useful example of confounding is provided by the arrangement of a three-factor experiment in blocks of four plots. If the factors be represented by a , b and c and we arrange the four treatment combinations

$$(1), ab, ac, bc,$$

in one block of each replication (randomizing within the block) and the other four combinations

$$a, b, c, abc,$$

in the other block; the contrast between these two sets of blocks is equivalent to the three-factor interaction $A.B.C$. Consequently all information on this interaction is lost, except the small amount arising from inter-block comparisons. It is easily seen, however, that block differences are eliminated from all the other interactions and from all the main effects, since each of these comparisons involves two plots with a plus and two plots with a minus sign from each block.

For reasons given in (9) it is best to arrange that neighbouring blocks are of unlike type, so that the blocks themselves form randomized pairs, each pair comprising a complete replication.

In order to illustrate the modifications that are necessary in the statistical analysis we will reconstruct the analysis of the potato experiment already given, on the supposition that it was arranged in this way and gave yields identical with those actually obtained. This will make clear the parallelism as well as the differences between the two analyses.* Actual examples of confounded experiments are given later in the paper.

4b. Statistical analysis.

The partition of the degrees of freedom in the analysis of variance is given in Table 12.

The formal analogy of this partition with that of split plot arrangements, discussed in Section 16a, should be noted. The blocks correspond to whole plots, arranged in blocks of 2, and the plots to sub-plots.

The appropriate error for testing $N.K.D$ is "within block pairs." Not only is this likely to be large, because it involves comparisons between whole blocks, but it is also very ill-determined, being based on only 3 degrees of freedom. Normally, therefore, the partition of the sum of squares "between blocks" is not performed, only the three components, blocks, treatments and error being calculated.

The steps of the whole calculation are as follows.

1. Calculate the sum of squares for blocks from the 8 block totals (given in Table 11).

*It may perhaps be well to emphasize that there is only one form of analysis appropriate to a given experimental arrangement. Thus it is not permissible, if it is found on analysis that the elimination of the sum of squares for blocks actually increases the estimate of the experimental error, as in the potato experiment just described, to omit to perform this elimination. The example which follows must therefore be taken as illustrating the statistical processes only.

TABLE 11. BLOCK TOTALS, *N.K.D* CONFOUNDED.

Ia	Ib	IIa	IIb	IIIa	IIIb	IVa	IVb
1163	1133	1157	1134	1168	1201	1209	1166

Blocks (b) contain *nkd*. Note that the sum of the b's less the sum of the a's equals [*N.K.D*].

2. Calculate the sum of squares for the unconfounded treatment comparisons by summing the squares of the relevant totals from Table 7. Check this (and Table 7) by calculating the sum of squares for all treatments (ignoring confounding) from the yields of the separate treatment combinations, and deducting the *N.K.D* component.

TABLE 12. ANALYSIS OF VARIANCE, *N.K.D* CONFOUNDED.

					D.F.	Sum of squares	Mean square
Between blocks	Between block pairs	3	774.1	258.0
	<i>N.K.D</i>	1	124.0	124.0
	Within block pairs	3	421.9	140.6
	Total	7	1320.0	188.6
Within blocks	Treatments*	6	458593.9	76432.3
	Error	18	6865.8	381.4
Total					31	466779.7	

*Main effects and interactions between two factors (see Table 9).

3. Calculate the error sum of squares by subtraction. The remainder of the analysis of variance and the tests of significance proceed as before.

4c. Presentation of results.

The presentation of the results requires slight modification, since any comparison involving *N.K.D* is affected by block differences. The best procedure is to divide the individual treatment combinations into two categories, according as they fall into blocks (a) or (b), arranging the results as in Table 13.

TABLE 13. YIELDS OF TREATMENT COMBINATIONS, *N.K.D* COMPLETELY CONFOUNDED.

	Blocks (a)				Blocks (b)				Mean
	(1)	<i>nk</i>	<i>nd</i>	<i>kd</i>	<i>n</i>	<i>k</i>	<i>d</i>	<i>nkd</i>	
Unadjusted	2.84	8.06	9.35	11.20	2.85	7.49	8.59	12.10	7.81
Assuming <i>N.K.D</i> = 0 ..	2.79	8.01	9.30	11.15	2.90	7.54	8.64	12.15	7.81

N.K.D will be omitted from the table of main effects and interactions.

If the table of individual treatment combinations is adjusted so that the mean of the first four components is equal to the mean of the second four by the addition of one half of the apparent value of *N.K.D*, here -0.05 tons, to each of the second four and the deduction of the same amount from each of the first four, this will eliminate block effects, at the cost of assuming that *N.K.D* is negligible. This procedure is not to be recommended as a general practice but is sometimes of value in the popular presentation of the results. All interactions between two factors, being unconfounded, can be presented by means of the ordinary two-way tables.

4d. Example of partial confounding.

Instead of confounding the three-factor interaction $A.B.C$ in every replication of a three-factor experiment the two-factor interactions may also be confounded in their turn. Thus the potato experiment might have been arranged in 8 blocks of 4 plots each, the interaction $N.K.D$ being confounded in the first pair, the interaction $N.K$ in the second pair, the interaction $N.D$ in the third and the interaction $K.D$ in the fourth. The treatments would then have had to be allotted to the pairs of blocks in the manner shown in Table 14.

TABLE 14. ARRANGEMENT OF TREATMENTS AND BLOCK TOTALS, PARTIAL CONFOUNDING.

Interaction confounded Block	$N.K.D$		$N.K$		$N.D$		$K.D$	
	Ia	Ib	IIa	IIb	IIIa	IIIb	IVa	IVb
Treatments	(1) nk nd kd	n k d nk	n k nd kd	(1) d nk nk	n d nk kd	(1) k nd nk	k d nk nd	(1) n kd nk
Total	1163	1133	1106	1185	1208	1161	1259	1116
Adjustment per plot..	-2.4	+2.4	+8.8	-8.8	-14.5	+14.5	+4.0	-4.0

If this procedure had been adopted, full information on the interaction $N.K.D$ would have been obtained from the block pairs II, III and IV, but no information would have been obtained from blocks I. Similarly, full information on $N.K$ would have been obtained from blocks I, III and IV, etc. Thus three-quarters of the information available on the main effects would be available on each of the interactions.

4e. Statistical analysis.

Certain modifications are required in the calculations of both the estimates of the interactions and the analysis of variance.

The general principle to be followed in cases of partial confounding is to estimate each partially confounded degree of freedom (or set of degrees of freedom) only from those blocks in which it is not confounded. Sums of squares are calculated from these estimates in the ordinary way, account being taken of the fact that they are based on a reduced number of plots. The sum of squares for blocks is computed from the block totals in the ordinary manner. The calculation will here run as follows.

The block totals must first be calculated. These are given in Table 14.

The totals for the interactions must be recalculated, omitting the blocks in which they are confounded. This can be done directly or by noting that, for instance, required total for $N.K = N.K$ total (Table 7) + total of block IIa - total of block IIb, or in the notation we shall adopt $[N.K]' = [N.K] + [IIa] - [IIb]$. This is the most expeditious method of calculation, but care must be taken with the signs. In our example

$$\begin{aligned}
 [N.K]' &= +105 + 1106 - 1185 = +26 \\
 [N.D]' &= +161 + 1208 - 1161 = +208 \\
 [K.D]' &= -669 + 1259 - 1116 = -526 \\
 [N.K.D]' &= -63 + 1163 - 1133 = -33
 \end{aligned}$$

The analysis of variance will now contain a degree of freedom for each interaction, since each can be estimated. The sum of squares for the interactions will be obtained by summing the squares of the above four numbers and dividing by 24, since each is the sum of plus or minus 24 yields. The sum of squares for the main effects will be identical with that already obtained in the unconfounded design. The sum of squares for blocks comes directly from the block totals. Finally the error sum of squares is obtained by subtraction. We thus obtain the analysis of variance shown in Table 15.

TABLE 15. ANALYSIS OF VARIANCE, PARTIAL CONFOUNDING.

			D.F.	Sum of squares	Mean square
Blocks	7	4499.0	642.7
Main effects	3	443453.1	147817.7
Interactions	4	13404.4	3351.1
Error	17	5423.2	319.0
Total	31	466779.7	

In this analysis it is not possible conveniently to subdivide the degrees of freedom for blocks, as was done when *N.K.D* was totally confounded.

The reader will notice that the estimates of error vary considerably in the three analyses, Tables 8, 12 and 15. This, however, does not indicate that the errors are different, since each is in fact an estimate of the same error. The variation is due entirely to random sampling variation resulting from the omission from the "error" of Table 8 of certain degrees of freedom, those "within block pairs" in Table 12, and others less easily isolated in Table 15.

The estimates of the interactions flow directly from the modified totals $[N.K]'$, etc. Since each comprises 24 plots the conversion factor must be that appropriate to the total of 12 plots, i.e. $\frac{60}{2240 \times 12}$ giving, in tons per acre, the values

$N.K = +0.06$, $N.D = +0.46$, $K.D = -1.17$, $N.K.D = -0.07$ values which, as should be the case, are not substantially different from those already found.

The estimate of the standard error of each of the totals $[N.K]'$, etc., is clearly

$$\sqrt{24 \times 319.0} = \pm 87.5$$

and this converted into tons per acre gives ± 0.195 . As before, the estimate of the standard error of the main effects will be $\sqrt{32 \times 319.0}$, giving ± 0.170 tons per acre.

Using the *t* test we find the 5% and 1% points for the interactions to be 0.411 and 0.565. Thus *K.D* is significant at the 1% level, and *N.D* now attains significance at the 5% level. This is an illustration of how, by considering part of the data only, effects which are insignificant when the whole of the data is taken into account, may by chance attain significance. Such tests are, of course, not valid, since they transgress the necessary condition that for any chosen effect in any given experiment there can be only one appropriate test of significance.

If we require the standard error of some function of the main effects and interactions, as for example the response to potash in the presence of dung :

$$K + K.D = + 3.80 - 1.17 = + 2.63$$

the ordinary rule of taking the square root of the sum of the squares of the standard errors of the two parts is applicable, since these parts are orthogonal and therefore in effect independent. The required standard error is therefore $\sqrt{(0.170^2 + 0.195^2)} = \pm 0.259$.

4f. *Presentation of results.*

In partially confounded experiments the ordinary table of the yields of the separate treatment combinations is misleading, since the values are affected by block differences, which may be very large. Since every interaction is determined, however, a table of adjusted yields may be computed. The experimenter will be well advised, wherever possible, to avoid presenting a comprehensive table of this nature, since it is troublesome to compute, and is also troublesome to interpret, since the various comparisons are not all of the same accuracy. If, however, such a table is required, it can be calculated from the main effects and interactions by the method already given. Tables embracing certain selected factors only are likely to be of more interest and utility, and can be similarly computed. Thus in the present experiment we might reasonably exhibit a two-way table of the combinations of dung and potash, similar to Table 3.

A useful check on the construction of tables of adjusted yields is provided by calculating the adjustments to the original yields necessary to eliminate block differences. Thus in our example the difference between blocks Ib and Ia should, if there were no block effects, give the interaction $N.K.D$. Since $[N.K.D]'$ contains 24 plots and blocks Ia and Ib together contain 8 plots the difference should be $\frac{1}{3} [N.K.D]' = -11$. Actually it is $1133 - 1163 = -30$. The adjustment per plot is therefore $\frac{1}{3} (30 - 11) = 2.4$, this being added to plots in Ib and subtracted from plots in Ia. The other adjustments shown in Table 14 are similarly computed. Thus the adjusted yield of combination nkd is

$$1807 + 2.4 - 8.8 + 14.5 - 4.0 = 1811.1$$

The reader will do well to satisfy himself that the use of these adjustments gives a table of adjusted yields which is identical with that obtained by reconstruction from the main effects and interactions.

5. SYSTEMS OF CONFOUNDING FOR $2 \times 2 \times 2 \times \dots$ DESIGNS.

In the last section the confounding of a single degree of freedom corresponding to the interaction between the three factors of a $2 \times 2 \times 2$ design was explained. We shall now consider the systems of confounding applicable to factorial designs involving four or more factors, each at two levels, i.e. designs of the form 2^n .

Clearly any single chosen degree of freedom for a main effect or interaction can be confounded, whatever the number of factors, for any such degree of freedom is derived from the contrast of one half of the treatment combinations with the other half, and it is therefore only necessary to assign these two groups

to the different blocks. If there are a large number of factors, however, a higher degree of confounding may be advisable. With 5 factors, for instance, there are 32 treatment combinations. If these are divided into groups of 8 in any way then the 3 degrees of freedom corresponding to the comparisons between the four groups will be confounded. The problem is so to choose the groups that these 3 degrees of freedom correspond to high-order interactions.

The possible solutions of this problem are provided by the following general rule :

If three degrees of freedom are to be confounded in a 2^n design any *two*, corresponding to main effects or interactions, may be chosen at will. The "generalized interaction" between these two degrees of freedom will then also be confounded. (By the generalized interaction between A.B.C and A.D, for example, is meant B.C.D, A being struck out as it occurs in both of the first two expressions.)

5a. Confounding with five factors.

In the case of the 2^5 design the main effects and interactions are those shown in Table 16.

TABLE 16. MAIN EFFECTS AND INTERACTIONS WITH FIVE FACTORS.

Main effects	Interactions between			
	two factors	three factors	four factors	five factors
A	A.B B.D	A.B.C A.D.E	A.B.C.D	A.B.C.D.E
B	A.C B.E	A.B.D B.C.D	A.B.C.E	
C	A.D C.D	A.B.E B.C.E	A.B.D.E	
D	A.E C.E	A.C.D B.D.E	A.C.D.E	
E	B.C D.E	A.C.E C.D.E	B.C.D.E	

If A.B.C.D.E is confounded, and also one of the interactions involving four factors, say B.C.D.E, then by the rule the main effect A is also confounded. The confounded set is thus

$$A; B.C.D.E; A.B.C.D.E$$

The only other type of set containing A.B.C.D.E is

$$A.B; C.D.E; A.B.C.D.E$$

There is also the type of set

$$A.B.C; A.D.E; B.C.D.E$$

This is the most useful of all, for no main effect or interaction between two factors is confounded. There are 15 such sets, for the factor corresponding to A can be chosen in 5 ways, and the remaining four factors can then be divided into two pairs in 3 ways.

The actual partition of the 32 treatment combinations into four blocks of 8, so that the chosen degrees of freedom are confounded, is effected by writing down the signs of any two of the three confounded degrees of freedom after the manner of Table 4, and allocating the four combinations + + , + - , - + , and - - so obtained to the four blocks.

By the device of partial confounding different sets may be confounded in the different replications. With 5 replications a balanced group of sets such as that given in Table 17 can be used, each of the interactions between three and four factors being confounded once and once only. In this case $\frac{1}{8}$ of the information (relative to that on the unconfounded degrees of freedom) will be sacrificed on these interactions.

TABLE 17. BALANCED GROUP OF SETS FOR 2^5 DESIGN IN BLOCKS OF 8 PLOTS.

A.B.C; A.D.E; B.C.D.E
 A.B.D; B.C.E; A.C.D.E
 A.C.E; B.C.D; A.B.D.E
 A.C.D; B.D.E; A.B.C.E
 A.B.E; C.D.E; A.B.C.D

The rule given above is capable of extended application. Thus if blocks of 4 plots are used in a 2^5 design and the interaction B.D is chosen, in addition to the first set of Table 17, the full set of 7 confounded interactions is

B.D; C.E; A.B.C; A.D.E; A.C.D; A.B.E; B.C.D.E

The eight combinations of signs arising from any three of these interactions (the third not being the generalized interaction of the other two) will give the partition into the eight blocks.

Balanced groups of 5 sets of this type also exist, one of these groups being that given in Table 18.

TABLE 18. BALANCED GROUP OF SETS FOR 2^5 DESIGN IN BLOCKS OF 4 PLOTS.

AB, CD, ACE, ADE, BCE, BDE, ABCD
 AC, DE, ABD, ABE, BCD, BCE, ACDE
 AD, BE, ABC, ACE, BCD, CDE, ABDE
 AE, BC, ABD, ACD, BDE, CDE, ABCE
 BD, CE, ABC, ABE, ACD, ADE, BCDE

5b. *Confounding with six factors.*

The confounding of experiments including six factors follows similar lines. With blocks of 16 treatments the most useful sets are those of the type

A.B.C.D; A.B.E.F; C.D.E.F

and with blocks of 8 treatments those of the type

A.C.E; B.D.E; B.C.F; A.D.F; A.B.C.D; A.B.E.F; C.D.E.F

With blocks of 4 treatments arrangements confounding 3 two-factor, 8 three-factor, 3 four-factor and the six-factor interaction are possible, and may be obtained by "interacting" on the sets of Table 18 with E.F, B.F, C.F, D.F and A.F respectively. A balanced group of sets will be thus attained. Balance is also possible in 5 replications with blocks of 16 treatments, but with blocks of 8 treatments, rather curiously, 10 replications are required for complete balance: with 5 replications and blocks of 8 plots one three-factor interaction must be confounded twice while another is not confounded at all.

5c. *Confounding with four factors in blocks of 4 plots.*

The best type of set for non-balanced arrangements is

A.B ; A.C.D ; B.C.D

but for complete balance this clearly demands 6 replications, and moreover $\frac{1}{2}$ the relative information on the three-factor interactions is lost. The alternative group of sets given in Table 19 gives balance in 4 replications, and sacrifices only $\frac{1}{4}$ of the relative information on the three-factor interactions and $\frac{1}{4}$ (instead of $\frac{1}{6}$) of the information on the two-factor interactions.

TABLE 19. 2^4 DESIGN.

A.B ; C.D ; A.B.C.D

A.C ; B.D ; A.B.C.D

A.D ; A.B.C ; B.C.D

B.C ; A.B.D ; A.C.D

There is the further group of 5 sets (Table 20) which confounds every degree of freedom once and therefore sacrifices $\frac{1}{5}$ of the relative information on every comparison. All comparisons are therefore of equal accuracy. This design depends structurally on the complete set of orthogonal 4×4 Latin squares.

TABLE 20. ALTERNATIVE 2^4 DESIGN.

A ; B ; A.B

C ; D ; C.D

A.C ; B.D ; A.B.C.D

A.D ; A.B.C ; B.C.D

B.C ; A.B.D ; A.C.D

5d. *General remarks.*

In agricultural field experiments in randomized blocks a very high degree of confounding is not usually advisable ; as a general rule the two-factor interactions should be left unconfounded. We have, however, thought it worth while to put on record the possible designs in blocks of 4 plots, both for the sake of completeness and because they may be found to be of use in other branches of biological experimentation where the block size is more definitely limited.

Balanced arrangements are particularly useful when the experimental material is such that a high degree of confounding is advisable, so that possibly important interactions are likely to be involved. In agricultural experiments the number of replications available is rarely great enough to attain balance in single experiments including large numbers of factors (though balance may be introduced in sets of experiments of similar design at different places). This does not preclude partial confounding, which should always be adopted when there is more than one replication and when a choice can be made between interactions of the same order, unless one set can be pronounced with certainty to be of no importance. Thus in the experiment on beans described in Section 7, in which the factors were spacing, dung, nitrogen, phosphate and potash, the three-factor interactions confounded with *S.D.P* and *S.N.K*. Had a second replication been available the three-factor interactions *D.P.K* and *S.N.P* might advantageously have been confounded in it. It is instructive to identify these sets with those given in general form in Table 17.

6. ESTIMATION OF ERROR FROM HIGH-ORDER INTERACTIONS.

A further difficulty which limits the number of factors that can be included in an experiment is the number of plots required. Thus with six factors 128 plots will be required for even two-fold replication.

If only a single replication is employed the experiment will not be capable of furnishing an estimate of error by the ordinary procedure of comparing replicates. There will, however, in large experiments be a number of interactions between three or more factors which may in many cases be confidently predicted to be small in comparison with the errors affecting them. If this is the case they will in effect themselves be estimates of experimental error. Thus, for example, in a 2^6 design no less than 22 of the 63 degrees of freedom for treatments correspond to interactions between four or more factors. If the experiment consists of a single replication and is arranged in blocks of 16 plots, three of these will be confounded with block differences. The remaining 19 may then be used as an estimate of experimental error.

It should be noted that even if some of these high-order interactions do happen, with some particular set of factors, to be appreciable, the experimenter is still in a much better position than he would have been had the interacting factor been omitted entirely from the design. For any particular interaction (except those which are confounded) which later results may indicate to be of importance can be isolated and examined. Moreover the criticism that the inclusion of an interaction of some magnitude in the estimate of experimental error will inflate that estimate does not carry much weight, since the true experimental error (as estimated between replicates of the same treatment combination) would not be applicable to the results of an experiment with the interacting factor held constant, if it were intended that these results should be treated as valid for *all* levels of the interacting factor.

This device of using only a single replication is particularly useful in agricultural field experiments. For it is well known that most of the effects which are being measured vary from year to year and place to place. A whole set of similar experiments, of moderate accuracy, conducted at different places over a series of years, is thus of far more value for practical purposes than a single large experiment of equivalent accuracy. The use of only a single replication enables experiments comprising a reasonable number of factors to be carried out on ordinary non-experimental farms, and thus very considerably adds to the resources of the experimenter.

7. AN EXPLORATORY EXPERIMENT ON BEANS.

As an example of the points discussed in the last two sections we will consider a 2^5 experiment on beans, conducted at Rothamsted in 1935.

The treatments consisted of all combinations of:

- (S) Spacing of rows: 18 ins. apart (s_0) or 24 ins. apart (s_1).
- (D) Dung: 10 tons per acre (d), or none.
- (N) Nitrochalk: 0.4 cwt. N per acre (n), or none.
- (P) Superphosphate: 0.6 cwt. P_2O_5 per acre (p), or none.
- (K) Muriate of potash: 1.0 cwt. K_2O per acre (k), or none.

The spacing was included to test the theory that the best effects of manures might be obtained with closely spaced rows.

The plan is shown in Table 21. The yields are given in the first column in Table 22.

TABLE 21. PLAN OF EXPERIMENT ON BEANS, AND BLOCK TOTALS.

Block III (555.2) Block IV (436.7)

s_0nk	s_0	s_0p	s_0npk
s_1np	s_1dn	s_1dnp	s_1n
s_0dp	s_1pk	s_0d	s_1k
s_1dk	s_0dnpk	s_0dnk	s_1dpk
s_1	s_1dp	s_0n	s_0k
s_1nk	s_0dk	s_1dnk	s_0dpk
s_0np	s_1dnpk	s_1p	s_0dnp
s_0dn	s_0pk	s_1d	s_1npk

Block I (412.3) Block II (481.0)

Only a single replication was used, giving 32 plots in all, each of $\frac{1}{8}$ acre, these being arranged in four blocks of 8 plots each. Examination will show that the following interactions are confounded:

Interaction		Contrast
$S.D.P$	I - II - III + IV
$S.N.K$	I + II - III - IV
$D.N.P.K$	I - II + III - IV

7a. Analysis.

The calculation of the main effects and interactions is given in Table 22, and the analysis of variance in Table 23.

The estimate of error is based on interactions between three or more factors. The computations follow exactly the same lines as those of the $2 \times 2 \times 2$ experiment. The sum of squares for treatments is obtained by dividing the sum of the squares of the totals of Table 22 corresponding to the main effects and two-factor interactions by 32 (there being no need to write down the individual squares), and the other two sums of squares are similarly obtained. A check is given in Table 22 for each of the columns (1) to (5), and a check of the whole set of calculations is provided by the total sum of squares, which is also calculated direct from the yields of the separate treatment combinations.

A further useful check is obtained if the block totals are noted when calculating the total sum of squares (as can conveniently be done by the method of Note 4 when, as often happens, the yields are tabulated by blocks). The confounded interactions can then be calculated directly from these block totals and compared with the values already obtained.

TABLE 22. CALCULATION OF MAIN EFFECTS AND INTERACTIONS, BEANS EXPERIMENT.

								Effect	
		Yield (o)	(1)	(2)	(3)	(4)	Effect (5)	cwt. per acre	
<i>sdnph</i>									
00000	66.5	102.7	232.2	461.3	881.8	1885.2	21.04	Mean
100	36.2	129.5	229.1	420.5	1003.4	- 125.0*	- 2.79	S
010	74.8	91.3	213.1	525.4	- 132.4	+251.2**	+5.61	D
110	54.7	137.8	207.4	478.0	+7.4	+80.6	+1.80	S.D
001	68.0	86.6	227.5	- 91.9	+156.6	+52.0	+1.16	N
101	23.3	126.5	297.9	- 40.5	+94.6	+53.0	+1.18	S.N
011	67.3	82.0	243.8	+5.8	+52.4	+82.4	+1.84	D.N
111	70.5	125.4	234.2	+1.6	+28.2	+31.8†		
00010	56.7	102.9	- 50.4	+73.3	- 8.8	- 88.2	- 1.97	P
100	29.9	124.6	- 41.5	+83.3	+60.8	+47.2	+1.05	S.P
010	76.7	131.7	- 53.7	+56.2	+75.8	- 7.8	- 0.17	D.P
110	49.8	166.2	+13.2	+38.4	- 22.8	- 187.2‡		
001	36.3	123.9	- 2.3	+58.1	+23.2	- 82.6	- 1.84	N.P
101	45.7	119.9	+8.1	- 5.7	+59.2	+14.4†		
011	60.8	95.9	+17.4	+75.8	+32.2	+17.4†		
111	64.6	138.3	- 15.8	- 47.6	- 0.4	- 10.0†		
00001	63.6	- 30.3	+26.8	- 3.1	- 40.8	+121.6*	+2.71	K
100	39.3	- 20.1	+46.5	- 5.7	- 47.4	+139.8*	+3.12	S.K
010	51.3	- 44.7	+39.9	+70.4	+51.4	- 62.0	- 1.38	D.K
110	73.3	+3.2	+43.4	- 9.6	- 4.2	- 24.2†	- 0.54	S.D.K
001	71.2	- 26.8	+21.7	+8.9	+10.0	+ 69.6	+1.55	N.K
101	60.5	- 26.9	+34.5	+66.9	- 17.8	- 98.6†		
011	73.7	+9.4	- 4.0	+10.4	- 63.8	+36.0†		
111	92.5	+3.8	+42.4	- 33.2	- 123.4	- 32.6†		
00011	49.6	- 24.3	+10.2	+19.7	- 2.6	- 6.6	- 0.15	P.K
100	74.3	+22.0	+47.9	+3.5	- 80.0	- 55.6†		
010	63.6	- 10.7	- 0.1	+12.8	+58.0	- 27.8†		
110	56.3	+18.8	- 5.6	+46.4	- 43.6	- 59.6†		
001	48.0	+24.7	+46.3	+37.7	- 16.2	- 77.4†		
101	47.9	- 7.3	+29.5	- 5.5	+33.6	- 101.6†		
011	77.0	- 0.1	- 32.0	- 16.8	- 43.2	+49.8†		
111	61.3	- 15.7	- 15.6	+16.4	+33.2	+76.4†		
							±51.2	±1.14	
Totals (for checks):									
1st half	{ Odds (a)	507.1	817.0	827.6	1164.0	1080.8	2109.6		
half	{ Evens (b)	374.7	1068.2	932.6	928.0	1230.4	- 95.2		
2nd half	{ Odds (c)	498.0	- 102.8	108.8	140.0	- 47.2	103.2		
half	{ Evens (d)	505.4	- 22.2	223.0	79.2	- 249.6	- 156.0		

Checks for column (1) $\begin{cases} a_1 + b_1 = + a_0 + b_0 + c_0 + d_0 \\ c_1 + d_1 = - a_0 + b_0 - c_0 + d_0 \end{cases}$
and similarly for the other columns.

*Significant (5 per cent. level), i.e. greater than 111 or 2.48 cwt.

**Significant (1 per cent. level), i.e. greater than 155 or 3.46 cwt.

†Used for estimate of error. ‡Confounded with blocks.

TABLE 23. ANALYSIS OF VARIANCE, BEANS EXPERIMENT.

	D.F.	Sum of squares	Mean square
Blocks	3	1476.43	492.14
Main effects and interactions between two factors	15	4921.20	328.08
Remainder	13	1066.64	82.05
Total	31	7464.27	

Spacing, dung, and potash have produced significant effects, and in addition the interaction between spacing and potash is significant. It is to be noted that the dung and spacing show a similar (though smaller and non-significant) interaction. The table (Table 24) including these three factors is therefore of interest. It is not affected by the confounding, and may be constructed either from the main effects and interactions or by taking the mean yields of the relevant sets of 4 plots.

TABLE 24. MEAN YIELDS, CWT. PER ACRE.

	(1)	k	d	dk
18 in. spacing	20.3	20.7	25.0	23.7
24 in. spacing	12.1	19.8	21.4	25.3

The experiment is not of high precision, being of only 32 plots, and having a high standard error per plot (beans have at Rothamsted proved a very variable crop), but in combination with other similar experiments it should provide useful information, and in itself affords an illustration of the importance of putting theories to experimental test, since the interaction between spacing and manures turned out to be the opposite of what had been expected.

7b. Gain in precision due to confounding.

It is clear that the arrangement in blocks has increased the precision, since the mean square for blocks is considerably greater than that for error. An estimate of the amount of this gain can be made by replacing the treatment mean square by the error mean square, and then calculating what the error would have been had there been no confounding. (This procedure assumes that the confounded interactions are negligible, and is, of course, subject to certain errors of estimation.)

The calculations are set out in full in Table 25. The estimate of the

TABLE 25. GAIN IN PRECISION DUE TO CONFOUNDING.

	D.F.	Sum of squares	Mean square
Blocks	3	1476.43	492.14
Within blocks	28	2297.40	82.05
Total	31	3773.83	121.74

error mean square for a block of 32 plots is 121.74, and the efficiency of an unconfounded arrangement is therefore $82.05/121.74$, or 67.4 per cent. The

reciprocal of this is 148.4 per cent. and the gain in information due to confounding is thus 48.4 per cent.

It should be noted that *if there is more than one replication, the whole of the sum of squares for blocks will not enter into the new estimate for error*; only those components which represent differences of blocks forming the same replication must be included.

8. CONFOUNDING IN LATIN SQUARE DESIGNS WITH FACTORS AT TWO LEVELS.

In a somewhat limited number of cases it is possible to adapt confounding to Latin square designs. Thus, for example, a 2^4 system involving 16 treatment combinations may be arranged in an 8×8 Latin square, there being four complete replications. Any one degree of freedom for a main effect or interaction may be confounded with rows (the rows being taken to represent blocks of 8 plots each), and at the same time another degree of freedom for a main effect or interaction may be confounded with columns. Alternatively partial confounding may be adopted, each of the 4 degrees of freedom for three-factor interactions being confounded in one of the four row-pairs, and the four-factor interaction being completely confounded in the four column-pairs. Three-quarters of the relative information will then be available on all three-factor interactions.

At the outset there is one point which should be emphasized. In order to obtain an unbiased estimate of error from a Latin square it is necessary to rearrange all rows in random order, and also all columns. Thus we are precluded from so arranging the experiment that the rows (or columns) forming each complete replication necessarily fall together in the field. This restriction is of importance in the types of design discussed in Section 16f and 16g, in which main effects such as varieties are confounded.

In spite of these limitations, such confounded Latin square designs as exist are of considerable interest, in view of the markedly greater precision of Latin squares as compared with randomized blocks in many types of agricultural field trials. We will therefore give examples which will illustrate the possibilities and limitations of this method of design. In this section we shall consider the various types which are applicable to sets of factors at two levels only. These must clearly utilize 4×4 and 8×8 squares. Further examples utilizing 6×6 and 9×9 squares will be given later.

8a. $2 \times 2 \times 2$ design in two 4×4 Latin squares.

Since we may arrange a 2^3 design in blocks of 4 plots in such a way as to confound any single degree of freedom, we may, in a single 4×4 square, totally confound any two interaction degrees of freedom, one with rows and one with columns, or alternatively we may partially confound two degrees with rows, and another two with columns. As in any case, however, at least two squares will be necessary to provide an adequate estimate of error, it is simpler, in cases in which partial confounding is required, to effect this by confounding the different degrees of freedom in different squares.

In experiments involving the three standard fertilizers there are various alternatives of possible utility. With two squares, for instance, *P.K* and *N.P.K*

may be confounded in both squares, or $N.P.K$ may be confounded with the columns of both, $P.K$ with the rows of one, and $N.P$ and $N.K$ partially with the rows of the other. With three squares $N.P.K$ may be confounded with the columns of all three squares, and $N.P$, $N.K$ and $P.K$ with the rows of one square each, thus obtaining $\frac{2}{3}$ the relative information on all two-factor interactions. Alternatively, if the two-factor interactions and the main effects are of equal interest, these may each be confounded in one half of one square, $N.P.K$ being confounded in all squares, giving $\frac{2}{3}$ the relative information on all effects except $N.P.K$.

The necessary designs are easily constructed by writing down the sets of treatment combinations that must fall together in the rows and the similar sets that must fall together in the columns. Thus to confound $P.K$ with the rows and $N.P.K$ with the columns the rows must consist of the two sets

(1)	n	pk	npk
p	k	np	nk

and the columns of the two sets

(1)	n
np	p
nk	k
pk	npk

This gives the following alternative squares (Table 26) with the first row and the first column in an assigned order :

TABLE 26.

(1)	n	pk	npk	(1)	n	pk	npk
np	p	nk	k	np	k	nk	p
nk	k	np	p	nk	p	np	k
pk	npk	(1)	n	pk	npk	(1)	n

For each square of the experiment one of the two squares may be selected at random, both the rows and columns being arranged in random order.

An alternative arrangement, which avoids confounding any two-factor interaction, is also worth noting. If the four treatment combinations (1), np , nk , pk , be arranged in a single 4×4 Latin square, and the other four combinations n , p , k , npk , in a second square, then the three-factor interaction $N.P.K$ will be identical with the comparison between the two squares. This arrangement has the defect that any differences in response to one of the factors, n say, in the two squares will give rise to an apparent interaction between the remaining factors p and k . This defect may be overcome, however, though with some probable loss of efficiency, by interlacing the two squares, one of each pair of columns (if there are eight columns) being assigned at random to the first square. Thus after randomization we might arrive at the arrangement given in Table 27.

TABLE 27.

(1)	k	pk	p	npk	np	nk	n
nk	npk	np	n	■	pk	(1)	p
pk	n	(1)	k	p	nk	np	npk
np	p	nk	npk	n	(1)	pk	■

The analysis will be conducted just as it would be if the squares were not interlaced, eliminating the rows as well as the columns of each square separately.

8b. Numerical example.

The above designs were superimposed on a uniformity trial on sugar beet conducted by Immer (17).

Table 28 shows the actual arrangement derived by randomization from Table 26 (the second square being selected in each case), and the yields of each plot ($\frac{1}{80}$ acre). *P.K* and *N.P.K* were confounded in both squares. The degree of freedom confounded with rows (also assigned at random from the above two) was *N.P.K* in the first square and *P.K* in the second.

TABLE 28. PLAN AND YIELDS IN LB.

<i>k</i>	<i>n</i>	<i>p</i>	<i>npk</i>	<i>p</i>	<i>np</i>	<i>nk</i>	<i>n</i>
542	587	583	576	549	562	576	569
<i>nk</i>	<i>pk</i>	<i>np</i>	(1)	<i>n</i>	<i>pk</i>	(1)	<i>npk</i>
629	615	634	594	637	623	643	629
<i>np</i>	(1)	<i>nk</i>	<i>pk</i>	<i>npk</i>	(1)	<i>pk</i>	<i>n</i>
562	596	624	627	639	628	645	651
<i>p</i>	<i>npk</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>nk</i>	<i>np</i>	<i>p</i>
604	638	609	634	615	586	605	618

The following estimates of the treatment effects (totals over 32 plots) were obtained :

$$N = +109, P = -11, K = +55; N.P = -147, N.K = -5.$$

The analysis of variance is given in Table 29.

TABLE 29. ANALYSIS OF VARIANCE, SEPARATE SQUARES.

		D.F.	Sum of squares	Mean square
Squares	1	457.5	457.5
Rows	6	20488.4	3414.7
Columns	6	2797.9	466.3
Treatments	5	1145.7	229.1
Error	13	3460.6	266.2
Total	31	28350.1	

The standard error of each of the above estimates is therefore ± 92.4 . No one of the effects is significant.

The analysis of variance appropriate to the arrangement in interlaced squares given in Table 27 is shown in Table 30.

TABLE 30. ANALYSIS OF VARIANCE, INTERLACED SQUARES.

		D.F.	Sum of squares	Mean square
Squares (= N.P.K)		1	442.5	442.5
Rows	6	18540.4	3090.1
Columns	6	2812.9	468.8
Treatments	6	2694.9	449.2
Error	12	3859.4	321.6
Total	31	28350.1	

It will be noted that in this example rows have been very effective in eliminating soil heterogeneity. Table 31 shows the mean squares obtained with squares and blocks of various types :

TABLE 31. EFFICIENCY OF VARIOUS ARRANGEMENTS.

					D.F.	Mean square		Relative efficiency
4 × 4 Latin squares	{ separate	18	..	255.9*	100.0
	{ interlaced	18	..	364.1*	70.3
Blocks of 4 plots	{ half-rows	24	..	308.5	82.9
	{ columns	24	..	1045.6	24.5
	{ 2 × 2 squares	24	..	940.7	27.2
Blocks of 8 plots	{ rows	28	..	407.6	62.8
	{ pairs of half-rows	28	..	867.7	29.5
	{ pairs of columns	28	..	949.1	27.0
Blocks of 16 plots	{ pairs of rows	30	..	829.5	30.8
	{ squares	30	..	929.8	27.5

*Treatments + error of Tables 29 and 30.

The major part of the soil heterogeneity lies in differences between rows, and consequently blocks along the rows are reasonably efficient. They are, however, a form of block which would not in practice be used unless prior information on the fertility differences of the field was available. The alternative forms of block, whether of 4 or 8 plots, have all efficiencies of less than 30 per cent. The arrangement in interlaced squares is somewhat less efficient than the arrangement in separate squares, but has served to eliminate the greater part of the variation due to rows.

It is not claimed that this example is typical of the average gain in efficiency that may be expected from the use of Latin squares instead of randomized blocks. It is, however, an excellent illustration of the power of Latin squares to deal with the types of soil heterogeneity met with in agriculture. In this connection it should be noted that if we have any type of experimental material which can be classified in two ways, with both of which variation is associated, then the elimination of both sources of variation simultaneously more than doubles the decrease in error variance over the average of that produced by the elimination of either source separately. Measured in terms of information per plot (which is equal to the reciprocal of the error variance per plot) the additional gain by the simultaneous elimination of both sources is even greater.

It is also to be remarked that if the variation associated with one type of classification is large, while that associated with a second type is negligible, the use of the second classification for blocks will always give a higher error than if the experiment were arranged wholly at random. In the present example the elimination of columns after eliminating rows has increased the information per plot from 82.9 to 100, whereas the elimination of columns before eliminating rows has decreased it from 27.5 to 24.5.

8c. *Arrangements for five and six factors in an 8×8 square.*

The arrangement of five and six factors in 4×4 squares is also possible if the confounding of some of the two-factor interactions is permitted, but the use of an 8×8 square appears more suitable, since all two-factor interactions can then be kept free from confounding.

In the case of five factors, groups of sets may be chosen from those shown in Table 17. If only a single square is available, partial confounding within the square may suitably be resorted to, four out of the five sets being confounded, two with rows and two with columns. In the square shown in Table 32 the first group of Table 17 is confounded in rows 1—4, the second in rows 5—8, the third in columns 1—4 and the fourth in columns 5—8, the fifth group being unconfounded. In this table the first of the pair of numbers gives the combination of the a , b , and c treatments, according to the scheme:

$$1 = (1), 2 = a, 3 = b, 4 = ab, 5 = c, 6 = ac, 7 = bc, 8 = abc,$$

and the second of the pair of numbers gives the d and e treatments, according to the scheme:

$$1 = (1), 2 = d, 3 = e, 4 = de.$$

Thus $72 = bcd$.

TABLE 32. 8×8 QUASI-LATIN SQUARE FOR FIVE FACTORS.

11	43	71	63	42	62	74	14
73	61	13	41	72	12	44	64
54	82	34	21	83	23	31	51
32	24	52	84	33	53	81	21
81	51	64	72	11	34	22	43
62	74	83	51	24	41	13	32
44	12	21	33	54	71	63	82
23	31	42	14	61	84	52	73

The analysis follows the ordinary lines, the partially confounded interactions being computed from the rows or columns in which they are unconfounded. There are thus 18 degrees of freedom for error. As before rows and columns must be completely randomized amongst themselves.

In the case of six factors the system of confounding will be of the type:
 Rows: A.C.E; A.D.F; B.D.E; B.C.F; A.B.C.D; A.B.E.F; C.D.E.F
 Columns: A.B.F; A.D.E; B.C.D; C.E.F; A.B.C.E; A.C.D.F; B.D.E.F
 The square shown Table 33 confounds these interactions. The second number now indicates one of the eight combinations of d , e and f .

TABLE 33. 8×8 QUASI-LATIN SQUARE FOR SIX FACTORS.

11	24	36	47	58	65	73	82
27	16	44	31	62	53	85	78
38	51	13	22	71	84	56	67
42	31	25	18	87	76	64	51
54	61	77	46	15	28	32	43
66	57	81	74	23	12	48	35
75	88	52	63	34	41	17	26
83	72	64	55	46	37	21	14

If 128 plots are available, a second square confounding a completely different set of three-factor interactions may be obtained from the above square by

changing *a* to *c*, *c* to *f*, *f* to *e*, and *e* to *a*. Two four-factor interactions will be confounded in both squares.

With only a single replication error will have to be estimated from high-order interactions. If all 12 unconfounded three-factor interactions are retained there will remain 16 degrees of freedom for error.

The actual factor which each letter is taken to represent in these designs must, of course, depend on the interest which attaches to the various interactions, the aim being to confound (as far as is possible) only those interactions which are likely to be of little importance.

The rows and columns of each square must be rearranged in random order for every experiment.

9. FACTORS AT MORE THAN TWO LEVELS.

In the preceding sections we have described factorial designs in which every factor is at two levels only. Many cases arise in practice, however, in which more than two levels of some or all of the factors are required. In all cases in which it is necessary to determine the optimal level of a factor, for instance, at least three levels are essential, and in factorial experiments in which varieties are included as one of the factors the use of three varieties rather than two is usually advisable.

When some or all of the factors are at more than two levels, part of the simplicity that attaches to factorial designs with factors at two levels only is lost. To the main effects of a factor at four levels, for instance, there will correspond 3 degrees of freedom, and similarly for all interactions involving this factor. The calculations required for the analysis of variance are consequently more complicated. Moreover the possibilities of confounding are much more restricted, and the designs which exist are less elegant and more troublesome statistically, particularly with factors at different numbers of levels.

In this section we will consider the modifications that are necessary in the statistical analysis when there is no confounding. In later sections the simpler types of confounding will be described.

9a. Two factors.

In a varietal and manuring experiment on oats (Rothamsted, 1931) four levels of nitrogen (0, 0.2, 0.4 and 0.6 cwt. per acre) were applied to each of three varieties, Victory, Golden Rain II and Marvellous. There were six replicates on $\frac{1}{10}$ acre plots. The total yields of each of the twelve treatment combinations are given in Table 34.

TABLE 34. VARIETAL AND MANURIAL EXPERIMENT ON OATS.

	Treatment totals ($\frac{1}{10}$ lb.)				Total
	n_0	n_1	n_2	n_3	
Victory	429	538	665	711	2343
Golden Rain II	480	591	688	749	2508
Marvellous	520	651	703	761	2635
Total	1429	1780	2056	2221	7486

Since there are twelve treatment combinations there must be 11 degrees of freedom for treatments. These can, as before, be divided into main effects and interactions.

There will be 3 degrees of freedom for the main effects of n , and 2 degrees of freedom for the varietal differences. This leaves 6 degrees of freedom for interactions. (Note that $6 = 3 \times 2$).

If (as is natural here) the main effects are defined as the average response to one factor at all levels of the other they will be derivable from the two sets of marginal totals of Table 34. The sums of squares corresponding to each set can be calculated in the ordinary manner from the sum of the squares of the deviations of these marginal totals, dividing by the number of plots in each. Thus the sum of squares for N is given by

$$\frac{1}{18} [1429^2 + 1780^2 + 2056^2 + 2221^2 - 18 \times 778336.06]$$

(Note the method, the most suitable for a calculating machine, of applying the correction for the mean. This correction, $7486^2/72$, should be calculated first and written down, as it is wanted repeatedly.)

The sum of squares for interactions cannot be conveniently calculated directly, and must therefore be obtained by subtraction from the total sum of squares for treatments. The full analysis is as follows (Table 35):

TABLE 35. PARTITION OF THE TREATMENT SUM OF SQUARES IN THE VARIETAL AND MANURIAL TRIAL.

		D.F.	Sum of squares	Mean square
Correction for mean	..		778336.06	
Nitrogen	3	20020.50	6673.50
Varieties	2	1786.36	893.18
Interactions	6	321.75	53.63
All treatments	11	22128.61	

There is no automatic check on this table, and all the computations must therefore be carefully checked.

It will be noted that the above computations are exactly analogous to those of the ordinary analysis of variance of a randomized block experiment. Nitrogen and varieties correspond to blocks and treatments, interactions to error, and all treatments to the total. The sums of squares and mean squares are divided by an additional factor 6 to allow for the fact that each value of Table 34 is the total of six plots.

We will discuss the layout and conclusions of this experiment in Section 16b.

9b. Three or more factors.

The extension of the above analysis to three or more factors follows on the same lines. In the case of three factors, a at 3 levels, b at 4 levels and c at 4 levels, for example, there will be 48 treatment combinations, and the partition of the degrees of freedom will be

A	2	A.B	6	
B	3	A.C	6	A.B.C 18.
C	3	B.C	6	

In order to calculate the sums of squares three two-way tables will be required, one between each of the three pairs of factors, the sums being taken over all the remaining factors. Each set of marginal totals occurs twice, thus providing useful checks on the construction of the table. These three tables will give the sums of squares for the main effects and interactions between two factors. The sum of squares for the interaction between all three factors can then be obtained by subtraction.

9c. Simplification when one of the factors is at two levels only.

If one of the factors is at two levels only the interactions of this factor with the others can be calculated directly by using the differences of the yields at the two levels of this factor for all combinations of the other factors, and analysing these in exactly the same manner as the totals of the yields at the two levels. In the case of two factors only the calculations can be arranged as in Table 36, which gives the total yields in pounds of the five replicates ($\frac{1}{80}$ acre plots) of an experiment on different proportions of oats and vetches in a forage mixture, both with and without nitrogen (Rothamsted, 1932).

TABLE 36. EXPERIMENT ON SEED MIXTURES AND NITROGEN.

	Seeding rates (lb. per acre).					Total
	200 oats No vetches	150 oats 50 vetches	100 oats 100 vetches	50 oats 150 vetches	No oats 200 vetches	
Without nitrogen	1405	1661	1788	1684	1342	7880
With nitrogen	1788	1979	2000	1792	1468	9027
Sum	3193	3640	3788	3476	2810	16907
Difference [$n - (1)$]	+ 383	+ 318	+ 212	+ 108	+ 126	+1147

The sum of squares for N is given by $1147^2/50$, and the sum of squares for the interactions is given by

$$\frac{1}{10}[383^2 + 318^2 + \dots - 229.4 \times 1147]$$

Table 37 shows the full analysis of variance.

TABLE 37. ANALYSIS OF VARIANCE OF EXPERIMENT ON SEED RATES.

	D.F.	Sum of squares	Mean square
Correction for mean ..		5716933.0	
Treatments	Seedings .. 4	60313.9	15078.5
	Nitrogen .. 1	26312.2	26312.2
	Interactions. . 4	5717.5	1429.4
	Total 9	92343.6	
Blocks 4		59601.9	14900.5
Error 36		28384.5	788.5
Total 49		180330.0	

Provided that the correction for the mean is computed twice, and that in calculating the interaction sum of squares the correction for the mean difference (equal to the sum of squares for N) is recomputed as shown, all the treatment sums of squares and the sums and differences of Table 36 are checked by computing the total sum of squares from the 10 values in the body of the table.

9d. *Procedure when two or more factors are at two levels only.*

The main effects and interactions involving the factors at two levels only may be computed by the method of Section 3 for each combination of the other factors. The analysis of these and their totals over the different levels of the other factors will give all the sums of squares required.

An example will make the procedure clear. The first three columns of Table 38 shows the total yields of the treatment combinations of a $3 \times 2 \times 2$ experiment on potatoes (Rothamsted, 1933). All combinations of

$$\left\{ \begin{array}{l} n_0 = \text{no artificial nitrogen} \\ n_1 = \text{sulphate of ammonia} \\ n_2 = \text{ammonia bicarbonate} \end{array} \right\} \times \left\{ \begin{array}{l} (1) = \text{no poultry manure} \\ m = \text{poultry manure} \end{array} \right\} \times \left\{ \begin{array}{l} (1) = \text{no super} \\ p = \text{super} \end{array} \right\}$$

were applied. There were three replicates on plots of $\frac{1}{85}$ acre. The arrangement was confounded in blocks of 6 plots, and is discussed in Section 13c.

TABLE 38. COMPUTATION OF MAIN EFFECTS AND INTERACTIONS OF A $3 \times 2 \times 2$ EXPERIMENT.

	Yields (lb.)								Effects				
	n_0	n_1	n_2	Total	n_0	n_1	n_2	Total	n_0	n_1	n_2	Total	
(1)	411	479	451	1341	855	1057	968	2880	2073	2361	2115	6549	Sum
p	444	578	517	1539	1218	1304	1147	3669	+129	+85	+121	+335	P
m	561	659	546	1766	+33	+99	+66	+198	+363	+247	+179	+789	M
mp	657	645	601	1903	+96	-14	+55	+137	+63	-113	-11	-61	P.M

The sums and differences of pairs of values in the first four columns are shown in the next four columns, and the sums and differences of these latter in the last four columns, which give the totals of the main effects and interaction of p and m for n_0 , n_1 and n_2 , and the total of all n . The total column forms a check on the operation at each stage.

The treatment sums of squares can now be calculated immediately. The correction for the mean is given by $6549^2/36$, the sum of squares for N by

$$\frac{1}{12} [2073^2 + 2361^2 + 2115^2 - 6549 \times 2183],$$

the sum of squares for P by $335^2/36$, the sum of squares for $P.N$ by

$$\frac{1}{12} [129^2 + 85^2 + 121^2 - 335 \times 111.66667]$$

and so on.

These sums of squares are set out in Table 39. The whole calculation is checked by calculating the treatment sum of squares from the individual treatment combinations.

In this particular experiment the degrees of freedom for M and $P.M.N$ were partially confounded, so the sums of squares for these degrees of freedom in Table 39 are not those that appear in the final analysis described later.

TABLE 39. PARTITION OF TREATMENT SUM OF SQUARES.

						D.F.	Sum of squares	Mean square
Correction for mean							1191372.2	
							<hr/>	
<i>N</i>	2	4034.0	2017.0
<i>P</i>	1	3117.4	3117.4
<i>P.N</i>	2	91.5	45.8
<i>M</i>	1	17292.2	17292.2
<i>M.N</i>	2	1442.7	721.4
<i>P.M</i>	1	103.4	103.4
<i>P.M.N</i>	2	1301.5	650.8
							<hr/>	
Total	11	27382.7	

If in the summary of the results two-way tables giving the yields of pairs of factors are required, that for the pairs of factors p and m can be derived immediately by conversion of the first total column of Table 38, while that for n and m can be derived by the conversion of the first two lines of the second set of four columns and the first line of the last set. Only that for the pair of factors n and p will require any fresh summations.

9e. *Two factors at three levels : formal subdivision of interactions in a 3×3 table.*

If the yield totals of the 9 treatment combinations are denoted by the numbers 1-9 according to the scheme of Table 40 :

TABLE 40. YIELD TOTALS.

	b_0	b_1	b_2
a_0	1	4	7
a_1	2	5	8
a_2	3	6	9

what may be called the two sets of *diagonal totals* of this table may be defined as

$$\begin{aligned}
 [I_1] &= 1 + 5 + 9 & [J_1] &= 1 + 6 + 8 \\
 [I_2] &= 2 + 6 + 7 & [J_2] &= 2 + 4 + 9 \\
 [I_3] &= 3 + 4 + 8 & [J_3] &= 3 + 5 + 7
 \end{aligned}$$

The four degrees of freedom for the interactions of a 3×3 table may be divided into two orthogonal pairs of degrees of freedom, for which the sums of squares are given by the appropriate fraction of the sums of the squares of the deviations of $[I]$ and of $[J]$ respectively, just as the sums of squares for the main effects are derived from $[A]$ and $[B]$. Equally a table of the mean yields of the treatment combinations can be constructed from a knowledge of $[A]$, $[B]$, $[I]$ and $[J]$, or the corresponding means. Thus, for example, with four replications,

$$\begin{aligned}
 a_1 b_2 &= \text{dev } A_1 + \text{dev } B_2 + \text{dev } I_3 + \text{dev } J_1 + \text{mean} \\
 &= \frac{1}{12} ([A_1] + [B_2] + [I_3] + [J_1]) - 3 \times \text{mean}.
 \end{aligned}$$

This formal subdivision provides a useful method of computation for the interactions of a single 3×3 table. The method is distinctly shorter than the

ordinary method of subtraction, since the whole computation then becomes self-checking. The analogous subdivision of the three-factor interactions in a $3 \times 3 \times 3$ design will be described when dealing with the confounding of this design.

The conventional numbering of the 9 treatment combinations of a pair of three-level factors given in Table 40 will be extensively used in subsequent pages. It should therefore be memorized. Note that the first factor is always written downwards.

9f. Example.

In an experiment on the manuring of meadow hay (Bakewell, 1935) the treatments (nothing, compost, and equivalent artificials) followed a two-year cycle, making 9 treatment combinations in all. The 1935 yields are given in Table 41. The marginal and diagonal totals are also shown in this table.

TABLE 41. YIELDS OF HAY IN 1935 IN LB. (TOTALS OF 4 PLOTS OF $\frac{1}{16}$ ACRE).

1933 and 1935 treatments	1932 and 1934 treatments			Total	Diagonal totals	
	Nil	Artificials	Compost		<i>I</i>	<i>J</i>
Nil	65.2	71.0	85.2	221.4	274.4	262.2
Artificials	104.2	101.0	112.2	317.4	274.2	283.4
Compost	94.5	84.8	108.2	287.5	277.7	280.7
Total	263.9	256.8	305.6	826.3		

The partition of the treatment sum of squares is shown in Table 42.

TABLE 42. PARTITION OF TREATMENT SUM OF SQUARES.

	D.F.	Sum of squares	Mean square
1932 and 1934 treatments ..	2	115.85	57.92
1933 and 1935 treatments ..	2	402.20	201.10
Interactions	4	22.83	5.71
All treatments	8	540.89	

Since the subdivision of the interaction degrees of freedom is formal, and does not correspond to any expected treatment effects, there is no point in calculating the two components of the sum of squares separately. The squares of all six diagonal totals are summed and 24 ($= 2 \times 12$) times the correction for the mean is deducted, before dividing by 12. The fact that the total of the three sums of squares equals the total sum of squares for treatments checks the whole computation. If the interaction sum of squares were not computed directly, every item would have to be checked.

The error mean square (24 d.f.) was 6.300. Thus there is no evidence of any interaction, and the effects of the fertilizers in the two years may be regarded as additive. The standard error of a marginal total is $\sqrt{12 \times 6.300}$ or ± 8.70 . Consequently the response to artificials applied in 1935 is significantly greater than to compost, but artificials applied in 1934 show no residual effect, whereas that of compost is significant and large.

10. CONFOUNDING WITH THREE AND FOUR FACTORS EACH AT THREE LEVELS.

Both $3 \times 3 \times 3$ and $3 \times 3 \times 3 \times 3$ experiments can be arranged in blocks of 9 plots or in 9×9 Latin squares, confounding only three-factor interactions. These designs are of considerable practical importance in agriculture, and we will therefore describe them in detail.

10a. $3 \times 3 \times 3$ designs in blocks of 9 plots.

There are 8 degrees of freedom for the three-factor interactions. These can be divided into four orthogonal pairs, each pair being given by the contrasts of the sums of three sets of nine plots each. The four groups of three sets are given in Table 43, being represented by the four letters W, X, Y, Z .*

TABLE 43. $3 \times 3 \times 3$ DESIGNS CONFOUNDING THREE-FACTOR INTERACTIONS.

Combination of first and second factors		W_1	W_2	W_3	X_1	X_2	X_3	Y_1	Y_2	Y_3	Z_1	Z_2	Z_3
		Level of third factor											
1	00	0	2	1	0	1	■	0	2	1	0	1	2
2	10	1	0	2	2	0	1	1	0	2	2	0	1
3	20	2	1	0	1	2	■	2	1	■	1	■	0
4	01	2	1	0	1	2	0	1	0	2	■	0	1
5	11	0	2	1	0	1	■	2	1	0	1	2	0
6	21	1	0	2	2	■	1	0	2	1	0	1	2
7	02	1	0	2	2	0	1	2	1	0	1	2	0
8	12	2	1	0	1	2	0	0	■	1	0	1	2
9	■	0	2	1	0	1	■	1	■	2	■	0	1

Examination of the table will show that every combination of each pair of factors occurs in each set of 9 plots, and consequently if these sets are arranged in blocks the main effects and two-factor interactions will be unconfounded.

If more than one replication is available it is best to use different groups for the different replications, thus partially confounding some or all of the three-factor interactions. If four replications are used complete balance is attained, and $\frac{2}{3}$ of the relative information will be available on all the three-factor interactions. Partial confounding introduces some additional complication into the computations, unless the partially confounded degrees of freedom are allowed to remain in the estimate of error, but the difficulties are not great if the method described below is systematically followed.

10b. Example of a $3 \times 3 \times 3$ design.

Table 44 gives the plan and yields of sugar in an experiment on sugar beet (Woburn, 1935) in which all combinations of three sowing dates, April 18th (d_0), May 9th (d_1), May 25th (d_2)†, three spacings of rows, 10 in. (s_0), 15 in. (s_1), 20 in. (s_2), and three levels of sulphate of ammonia, nothing (n_0), 0.3 cwt. N per acre (n_1), and 0.6 cwt. N per acre (n_2), were included. The experiment was

*These groups have previously been numbered I, II, III and IV in various orders, but no consistent notation has been established.

†An earlier sowing, March 14th, failed and d_2 replaced this.

arranged in 6 blocks of 9 plots each. Since after rejection of edge rows the plots of the three spacings were of different area the yields have been converted to cwt. per acre before analysis.*

TABLE 44. PLAN AND YIELDS OF SUGAR (CWT. PER ACRE).

Y_2 425.8	62	52.2	90	31.3	Z_2 305.5
	51	52.7	20	36.4	
	20	47.8	11	39.4	
	90	35.2	61	35.3	
	82	45.4	32	29.9	
	40	44.6	81	34.4	
	31	46.0	40	33.3	
	71	51.4	52	33.6	
	12	50.5	72	31.9	
Y_1 439.9	41	49.7	12	33.6	Z_3 314.3
	10	47.8	82	31.4	
	32	44.1	70	25.7	
	21	52.5	50	33.0	
	52	49.3	41	36.6	
	91	46.2	62	41.4	
	60	47.1	91	37.6	
	80	47.2	30	33.2	
	72	56.0	21	41.8	
Y_3 359.0	42	50.9	80	32.4	Z_1 317.8
	61	38.2	92	37.7	
	22	43.0	10	39.4	
	81	36.5	22	43.1	
	30	38.0	71	34.9	
	11	45.7	51	34.2	
	50	37.1	42	36.0	
	92	34.2	60	33.5	
	70	35.4	31	26.6	

The combination of the first two factors, d and s , on each plot is given by the first figure, and the level of the third factor, n , by the second figure.

The various steps in the analysis of an experiment of this type are as follows. The order given should be adhered to, so that errors may be detected before the erroneous values are used in extensive further calculations.

1. Identify the blocks with the groups and sets given in Table 43, or check the numbering if this is given.

2. Set out the totals of the separate treatment combinations in the order shown in Table 45 (first factor down, second and third across, with third uppermost). This should be done even if there is only a single replication.

*This accounts for slight differences between the results given here and those in the Rothamsted Report.

TABLE 45. YIELDS OF SEPARATE TREATMENT COMBINATIONS.

	No			No			No		
	So	Si	Se	So	Si	Se	So	Si	Se
d_0	87.2	77.9	61.1	85.1	86.3	86.3	84.1	86.9	87.9
d_1	84.2	70.1	79.6	94.3	86.9	70.9	86.1	82.9	76.8
d_2	71.2	80.6	66.5	72.6	73.5	83.8	74.0	93.6	71.9

3. Calculate the total sum of squares of all the yields of Table 44, the correction for the mean (which should be checked), the sum of squares for blocks, and the total sum of squares for treatments from Table 45. The block totals are obtained in the course of this calculation, together with a check on the total and on the formation of Table 45 (see Note 4).

4. Calculate the five 3×3 tables given in Table 46a. The first three require no comment. The last two give the diagonal totals $[I]$ and $[J]$ for the 3×3 tables for each level of n of Table 45. Marginal totals need not be taken out at this stage.

TABLE 46. CALCULATION OF MAIN EFFECTS AND INTERACTIONS.

(a) Two-way tables.

	s_0	s_1	s_2	
d_0	256.4	251.1	235.3	742.8
d_1	264.6	239.9	227.3	731.8
d_2	217.8	247.7	222.2	687.7
	n_0	n_1	n_2	
d_0	226.2	257.7	258.9	
d_1	233.9	252.1	245.8	
d_2	218.3	229.9	239.5	
s_0	242.6	252.0	244.2	738.8
s_1	228.6	246.7	263.4	738.7
s_2	207.2	241.0	236.6	684.8
I_1	223.8	255.8	238.9	
I_2	225.9	254.1	267.6	
I_3	228.7	229.8	237.7	
J_1	247.4	229.5	254.5	
J_2	228.6	264.4	244.9	
J_3	202.4	245.8	244.8	
	678.4	739.7	744.2	2162.3

(b) Three-factor interactions.

	1	2	3	Total
[W]	715.6	694.6	752.1	2162.3
[X]	721.2	719.4	721.7	
[Y]	756.6	728.9	676.8	
Blocks	439.9	425.8	359.0	1224.7
[Y]'	316.7	303.1	317.8	937.6
				312.53
[Z]	738.1	702.9	721.3	
Blocks	317.8	305.5	314.3	937.6
[Z]'	420.3	397.4	407.0	1224.7
				408.23

Standard errors. Totals of 6: ± 8.97 ; totals of 18: ± 15.54

5. Calculate the diagonal totals of the $[I]$ and $[J]$ tables in the proper order ($[I]$ and $[J]$ of the $[I]$ table, $[I]$ and $[J]$ of the $[J]$ table). These give (ignoring the confounding) the totals $[W]$, $[X]$, $[Y]$, $[Z]$ for the four pairs of three-factor interactions, and are shown in Table 46b.

6. Calculate the sums of squares corresponding to the nine values in each of the first three tables of Table 46a. These are shown in Table 47. The first table, for instance, gives the sum of D , S , and $D.S$. One set of marginal totals of each of these three tables may be obtained in the course of this calculation.

TABLE 47. AUXILIARY TABLE OF TREATMENT SUMS OF SQUARES (IGNORING CONFOUNDING).

Correction for mean	86584.10
Correction for working mean ($\frac{100}{9}$)	2430.76
$D, S, D.S$	341.52
$D, N, D.N$	275.13
$S, N, S.N$	329.77
D	94.47
S	107.80
N	150.14
$D.S.N$	Unconfounded W, X			..	94.22
	Partially confounded Y, Z			..	216.84
All treatments	905.06

7. Calculate the sums of squares for the main effects from these marginal totals (checking that the total of each set is correct), and enter these in Table 47.

8. Calculate the sums of squares for the four pairs of degrees of freedom for the three-factor interactions, keeping separate the unconfounded and partially confounded degrees of freedom, and enter these in Table 47.

9. Subtract the sums of squares for main effects from the sum of all the other treatment items of Table 47. This should give the total treatment sum of squares and assures the correctness of all of the preceding calculations which involve treatments.

10. Check the sum of squares for blocks and the total sum of squares.

If there were no confounding, or if one pair of degrees of freedom were completely confounded, the final analysis of variance table could now be immediately prepared. With partial confounding, however, the following additional steps are necessary.

11. Enter the block totals corresponding to the confounded pairs of degrees of freedom in the proper order in Table 46b, subtract these from the full totals, $[Y]$ and $[Z]$, thus obtaining the totals $[Y]'$ and $[Z]'$, which include only those blocks in which Y and Z respectively are not confounded. (If there is any doubt about this process, check one or more of the values by direct totalling over the blocks in which the degrees of freedom concerned are not confounded.) Calculate the sum of squares from these new totals and enter in Table 47. (Note that each set has a different total and therefore a different correction for the mean, and that a new divisor, here 9, is required, since only 9 plots are included in each total.) *The whole of this calculation must be checked, particular attention being paid to seeing that the block totals are entered in their correct places.*

12. Construct the final analysis of variance table shown in Table 48.

TABLE 48. ANALYSIS OF VARIANCE.

		D.F.	Sum of squares	Mean square	<i>z</i>
Correction for mean		86584.10		
Blocks	5	1950.38	390.08	
<i>D</i>	■	94.47	47.24	0.629*
<i>S</i>	2	107.80	53.90	0.695*
<i>N</i>	■	150.14	75.07	0.861*
<i>D.S</i>	4	139.25	34.81	0.477
<i>D.N</i>	4	30.52	7.63	
<i>S.N</i>	4	71.83	17.96	0.146
<i>D.S.N</i>	Unconfounded	4	94.22	17.31	0.127
	Partially confounded	4	44.29		
Error	22	295.29	13.42	
Total	53	2978.19		

13. Construct the various summaries of results. Tables for main effects and two-factor interactions and their standard errors can be obtained directly by conversion of the first three tables of Table 46. The conversion factor is here $\frac{1}{8}$.

In this experiment the reduction in error variance by the arrangement in blocks is very large. Although much of this reduction results from the difference between the two replications, the further reduction due to the use of blocks of 9 instead of 27, made possible by confounding, is also substantial, the gain in information, estimated by the method of Section 7*b*, being 53.1 per cent.

10c. Adjusted yields of three-factor combinations.

Under ordinary circumstances it will not be necessary to construct any table including all three factors, but should this be required it may best be done in two stages:

- assuming the three-factor interactions to be negligible;
- introducing correcting terms for these interactions.

The general rule for obtaining any value of stage (a) is to take the sum of the appropriate values of the *converted* two-way tables representing two-factor interactions, deducting the corresponding marginal *means* the number less one of times they are involved (i.e. once with three factors, twice with four factors, etc.) and adding the requisite multiple of the general mean. Thus in the above example: $d_0s_0n_0 = 42.73 + 37.70 + 40.43 - 41.27 - 41.04 - 37.69 + 40.04 = 40.90$, 42.73 being $\frac{1}{8}$ of 256.4 and 41.27 being $\frac{1}{8}$ of 742.8, etc.

The correcting terms for the three-factor interactions are immediately obtainable from Table 46*b* by multiplying [*W*] and [*X*] by the conversion factor for 18 plots (here $\frac{1}{18}$) and [*Y*]' and [*Z*]' by the conversion factor for 9 plots (here $\frac{1}{9}$). Since $d_0s_0n_0$ occurs in *W*₁, *X*₁, *Y*₁ and *Z*₁, the corrected value is

$d_0s_0n_0 = 40.90 + 39.76 + 40.07 + 35.19 + 46.70 - 4 \times 40.04 = 42.46$,
the mean of the means of *Y*' and *Z*' being equal to the general mean.

Alternatively corrections may be applied to the individual plot yields so as to eliminate the block effects, as in Section 4f. These are derived from Table 46b, that for block Y_1 , for instance, being

$$\frac{1}{9} (316.7 - 312.53 - 439.9 + 408.23) = -3.06.$$

Similarly that for block Z_1 is $+0.76$, and consequently the adjusted yield of $d_0s_0n_0$ is (from Table 45)

$$\frac{1}{2} (87.2 - 3.06 + 0.76) = 42.45.$$

To prevent the accumulation of small errors and facilitate checking it is best to retain an additional figure in this calculation, as shown. When the whole table is required the computation can be shortened in various ways, the details of which may be left to the reader.

The standard errors of the various differences can be obtained by considering which of the interaction effects W , X , Y and Z are involved, remembering that each difference is made up of the sum of 9 components, representing main effects, and two- and three-factor interactions. Thus $d_1s_1n_1$ and $d_0s_0n_0$ occur in the same Z set, but in different W , X , and Y sets. The relative information on Y is $\frac{1}{2}$, and consequently the variance as ordinarily calculated must be increased in the ratio

$$(8 \cdot \frac{1}{2} + 1 \cdot \frac{2}{3}) : 1 = 10 : 9.$$

Similarly $d_1s_0n_0$ and $d_0s_0n_0$ occur in different W , X , Y and Z sets, so that the variance of their difference must be increased in the ratio $11 : 9$. Had there been four replications, with $\frac{2}{3}$ information on W , X , Y and Z , the ratios would have been $10 : 9$ and $31 : 27$ respectively.

The calculation of separate components of the three-factor interactions is discussed in the next section.

10d. $3 \times 3 \times 3 \times 3$ designs in blocks of 9 plots.

Designs with four factors (but not more) at three levels can be arranged in blocks of 9 plots in a similar manner to designs with three factors, confounding only three-factor interactions. Consequently, if 81 plots are available, the possibility of including an additional factor should always be borne in mind, since this entails no loss of accuracy owing to increase in block size and little additional complication in the computations.

There are 32 degrees of freedom for three-factor interactions. These can be divided in various ways into 4 groups of 8 degrees of freedom each, in such a manner that each group of 8 degrees of freedom is given by the contrasts of 9 sets of 9 treatment combinations. One such group of sets is shown in Table 49. In this table the combinations of the third and fourth factors are also represented by the numbers 1-9. Thus the fourth combination of the second set of the first grouping has the number 47, which represents the combination $a_0b_1c_0d_2$. The table is used in an exactly similar manner to Table 43.

The analysis of variance follows the same lines as that of the $3 \times 3 \times 3$ design. In experiments with a single replication, however, it is scarcely worth while computing every item of the analysis of variance separately. The sums

of squares for the main effects and two-factor interactions may be calculated from two-way tables in the ordinary manner. The three-factor interactions between any set of three factors which are judged to be of interest may also be eliminated from the estimate of error if desired. A pair of degrees of freedom out of any such set of 8 is confounded with blocks.

TABLE 49. SET OF 3^4 DESIGNS CONFOUNDING THREE-FACTOR INTERACTIONS.

Combination of first and second factors		Combination of third and fourth factors									
		I					II				
1	00	1	5	9	8	3	4	6	7	2	
2	10	5	9	1	3	4	8	7	2	6	
3	20	9	1	5	4	8	3	2	6	7	
4	01	6	7	2	1	5	9	8	3	4	
5	11	7	2	6	5	9	1	3	4	8	
6	21	2	6	7	9	1	5	4	8	3	
7	02	8	3	4	6	7	2	1	5	9	
8	12	3	4	8	7	2	6	5	9	1	
9	22	4	8	3	2	6	7	9	1	5	
Confounded degrees of freedom		<i>A.B.C</i> (<i>W</i>), <i>A.B.D</i> (<i>Y</i>) <i>A.C.D</i> (<i>Z</i>), <i>B.C.D</i> (<i>X</i>)					<i>A.B.C</i> (<i>X</i>), <i>A.B.D</i> (<i>Z</i>) <i>A.C.D</i> (<i>W</i>), <i>B.C.D</i> (<i>Y</i>)				
		III					IV				
1	00	1	6	8	9	2	4	5	7	3	
2	10	6	8	1	2	4	9	7	3	5	
3	20	8	1	6	4	9	2	3	5	7	
4	01	9	2	4	5	7	3	1	6	8	
5	11	2	4	9	7	3	5	6	8	1	
6	21	4	9	2	3	5	7	8	1	6	
7	02	5	7	3	1	6	8	9	2	4	
8	12	7	3	5	6	8	1	2	4	9	
9	22	3	5	7	8	1	6	4	9	2	
Confounded degrees of freedom		<i>A.B.C</i> (<i>Z</i>), <i>A.B.D</i> (<i>W</i>) <i>A.C.D</i> (<i>X</i>), <i>B.C.D</i> (<i>W</i>)					<i>A.B.C</i> (<i>Y</i>), <i>A.B.D</i> (<i>X</i>) <i>A.C.D</i> (<i>Y</i>), <i>B.C.D</i> (<i>Z</i>)				

If the totals of the blocks of any grouping (taken in the order shown) are arranged in a two-way table in the standard order (Table 40), then the column totals give the confounded degrees of freedom from *B.C.D*, the row totals *A.C.D*, the *I* totals *A.B.D*, and the *J* totals *A.B.C*. The actual pairs confounded are given in Table 49; they can also be easily identified by determining which of the sets of totals, [*W*], [*X*], [*Y*] or [*Z*], for the factors concerned contains whole blocks in each total instead of three plots from each block. If no three-factor interactions are eliminated there will be 40 degrees of freedom for error; if all are eliminated there will be 16 degrees of freedom for error.

10e. 3^3 and 3^4 designs in quasi-Latin squares.

It follows from the arrangements already given for confounding in randomized blocks, that both 3^3 and 3^4 designs can be arranged in 9×9

quasi-Latin squares, only three-factor interactions being confounded. Arrangements of this type are shown in Tables 50 and 51. Rows and columns must be randomized as usual. Partial confounding could be adopted in the 3^3 design but is scarcely worth while in a single square, since $\frac{2}{3}$ the relative information must be sacrificed on two of the pairs of degrees of freedom.

TABLE 50 $3 \times 3 \times 3$ DESIGN IN A 9×9 QUASI-LATIN SQUARE.

10	21	32	41	52	60	72	80	91
21	32	10	52	60	41	91	72	80
32	10	21	60	41	52	80	91	72
42	61	50	92	81	70	30	11	22
50	42	61	70	92	81	11	22	30
61	50	42	81	70	92	22	30	11
71	90	82	12	31	20	40	62	51
82	71	90	20	12	31	62	51	40
90	82	71	31	20	12	51	40	62

Confounded degrees of freedom: rows, Y ; columns, W (Table 43).

TABLE 51. 3^4 DESIGN IN A 9×9 QUASI-LATIN SQUARE.

11	29	35	48	54	63	76	82	97
28	34	13	56	62	47	81	99	75
36	12	27	61	49	55	98	74	83
45	51	69	73	88	94	17	26	32
53	68	44	87	96	72	25	31	19
67	46	52	95	71	89	33	18	24
79	85	91	14	23	38	42	57	66
84	93	78	22	37	16	59	65	41
92	77	86	39	15	21	64	43	58

Confounded degrees of freedom: rows, II ; columns, IV (Table 49).

10f. Extension to 3^n in blocks of 3^{n-1} or 3^{n-2} .

If in Table 43 we replace each level the third factor by a set of three combinations of a third and a fourth factors, such that, in the previous notation, $0 = 1 + 5 + 9$, $1 = 2 + 6 + 7$, $2 = 3 + 4 + 8$ (the I sets), then the contrast of W_1 , W_2 and W_3 , etc., will represent a pair of degrees of freedom from the four-factor interactions $A.B.C.D$. If the J sets are used, then another pair of degrees of freedom will be obtained. Thus all the 16 degrees of freedom will be obtained in pairs. We are consequently provided with a set of designs for confounding a 3^4 design in blocks of 27 plots.

The process may be continued indefinitely, and a similar process may be applied to the 3^4 designs in blocks of 9 plots to give 3^5 designs in blocks of 27 plots, etc.

11. THE SUBDIVISION OF SETS OF DEGREES OF FREEDOM.

11a. Subdivision of main effects.

If the response to a fertilizer is proportional to the amount of the fertilizer present, i.e. if the response curve is a straight line, and if the fertilizer is applied at three levels, equally spaced, the response per unit dressing will be estimated

from the difference of the two extreme values. Moreover in such a case the yield of the central dressing will be equal to the mean of the yields of the two extreme dressings, except for experimental error, and consequently the observed difference of these two quantities may therefore be taken as a measure of the curvature of the response curve.

We may thus divide the two degrees of freedom for a fertilizer, n say, at three levels into two single degrees of freedom, one representing the average or *linear* component of the response and the other the curvature. These quantities may be denoted by N' and N'' , defined as

$$N' = n_2 - n_0$$

$$N'' = n_2 - 2n_1 + n_0$$

N' is therefore the response to the double dressing, and N'' the difference between the responses to the second and to the first dressing.*

The sums of squares corresponding to N' and N'' are given by

$$\frac{1}{2n} [N']^2 \text{ and } \frac{1}{6n} [N'']^2$$

respectively ($6 = 1^2 + 2^2 + 1^2$), where n is the number of plots contributing to $[n_0]$, etc. The standard errors are $\sqrt{2/n}$ and $\sqrt{6/n}$ times the standard error of a single plot. The two degrees of freedom are orthogonal, and consequently the two sums of squares total to the sum of squares for the two degrees of freedom.

If the response is substantially linear over the range investigated the sum of squares for N' will be much greater than that for N'' , and it may well be that N' attains significance although the sum of squares for the two degrees of freedom fails to do so, owing to the diluting effect of N'' . The test of N' alone is always legitimate, and should be made when the two degrees together fail to attain significance and inspection of the results indicates that N' may do so. The experimenter who confines his attention to the two degrees together is in fact penalizing himself by the very act of including in the experiment the intermediate level. In practice it is not necessary to calculate the separate sums of squares, since both N' and N'' can be immediately tested by the t test, using the final summary of results.

Thus in the example just given the mean square for sowings, 47.24 (2 d.f.), is only just significant at the 5% point, but the major portion, 84.33, of the corresponding sum of squares, 94.47, is attributable to the linear component D' , which is thus clearly significant. The actual difference D' is -3.06 cwt. per acre, and its standard error is ± 1.22 , giving $t = 2.51$. On the other hand the curvature D'' , which has a value -1.84 cwt. per acre, and a standard error of ± 2.11 , does not approach significance. The corresponding sum of squares is 10.15, giving the correct total.

The reader will find it instructive to examine the response curves for spacing and nitrogen in a similar manner. Although all the curvatures are in the direction that might be expected no one of them is significant. This illustrates the high precision that is necessary to determine the curvature of the response curve at all accurately.

*These quantities are represented by N_1 and N_2 Fisher's *Design of Experiments*. In view of the wide use of suffixes to indicate levels of a factor, however, we have thought it better to use dashes.

Similar divisions can be made when other types of treatment are involved. Thus in the experiment given in Table 41 the two degrees of freedom for 1933 and 1935 treatments might be divided into two single degrees of freedom, one representing the response to fertilizers, i.e. the mean of artificials and compost, and the other the difference between artificials and compost. Note, however, that if the single degrees of freedom were chosen to represent the response to artificials and the response to compost, these would not be orthogonal, and consequently the corresponding sums of squares, although each would in itself give rise to a z test of significance identical with the t test, would not add up to the total sum of squares for this set of treatments. There is no reason why the separate comparisons considered should always correspond to orthogonal degrees of freedom, but this will most frequently be the case in well designed experiments.

Sets of three or more degrees of freedom can be divided in a similar manner. There are many possible alternatives, which we have not the space to discuss here. The point to remember about all such subdivisions is that to be useful they must correspond to some reasonable simplification of the treatment effects, e.g. that forms of nitrogen are equivalent, that the response curve to a fertilizer can be reasonably represented by a straight line, or a second degree curve, etc. Whether such simplifications are in fact contradicted by the data can then be rigorously tested.

11b. Subdivision of interactions.

Corresponding to any given subdivision of the degrees of freedom for the main effects of a factor, there exists a corresponding subdivision of the associated interaction degrees of freedom. Thus in the previous example the four degrees of freedom for the interactions between sowing dates and spacings may be subdivided into the interaction of the linear responses $D'.S'$, the interactions of each linear response with the other curvature, $D''.S'$ and $D'.S''$, and the interaction of the two curvatures, $D''.S''$. $D'.S'$, for example, indicates the linear component in the change, as s varies, of the linear response to d , or alternatively to s as d varies.

The quantitative expressions for these interactions present no difficulty. Thus the linear response to d at the level s_0 of s is $d_2s_0 - d_0s_0$ and that at the level s_2 is $d_2s_2 - d_0s_2$. The difference of these

$$d_2s_2 - d_0s_2 - d_2s_0 + d_0s_0$$

gives the change in the linear response to d . Following our previous practice, we shall introduce the factor $\frac{1}{2}$, so that symbolically

$$D'.S' = \frac{1}{2}(d_2 - d_0)(s_2 - s_0)$$

Equally

$$\begin{aligned} D''.S' &= \frac{1}{2}(d_2 - 2d_1 + d_0)(s_2 - s_0) \\ D'.S'' &= \frac{1}{2}(d_2 - d_0)(s_2 - 2s_1 + s_0) \\ D''.S'' &= \frac{1}{2}(d_2 - 2d_1 + d_0)(s_2 - 2s_1 + s_0) \end{aligned}$$

The multipliers of the yield totals and the divisors required to give the sums of squares in the analysis of variance are given in tabular form in Table 52,

TABLE 52. EXPRESSIONS FOR INTERACTIONS OF A 3×3 TABLE.

	$A'.B'$			$A''.B'$			$A'.B''$			$A''.B''$		
	b_0	b_1	b_2	b_0	b_1	b_2	b_0	b_1	b_2	b_0	b_1	b_2
a_0	+1	0	-1	-1	0	+1	-1	+2	-1	+1	-2	+1
a_1	0	0	0	+2	0	-2	0	0	0	-2	+4	-2
a_2	-1	0	+1	-1	0	+1	+1	-2	+1	+1	-2	+1
Divisor	$4n$			$12n$			$12n$			$36n$		

n being the number of plots included in each total of the 3×3 table. As usual the divisors required to give the interactions in units of a single plot yield are *one-half* the above divisors, and the multipliers of the error mean square required to give the error variances of the *totals* are *equal* to the above divisors.

IIIc. Example.

Applying the above multipliers to the d and s table of the previous example, we obtain the results of Table 53.

TABLE 53. NUMERICAL VALUES OF INTERACTIONS.

Interaction	Total	cwt. per acre	Sum of squares
$D'.S'$	+25.5 \pm 17.9	+2.12 \pm 1.48	27.09
$D''.S'$	+57.9 \pm 31.1	+1.61 \pm 0.86	46.56
$D'.S''$	-44.9 \pm 31.1	-1.25 \pm 0.86	28.00
$D''.S''$	-90.1 \pm 53.8	-0.83 \pm 0.50	37.58
			<u>139.23</u>

A systematic method of arriving at the above totals, and also the totals for the corresponding main effects, is shown in Table 54. In the first three

TABLE 54. COMPUTATION OF MAIN EFFECTS AND INTERACTIONS OF A 3×3 TABLE.

(1)			(2)			Key		
738.8	738.7	684.8	2162.3	-54.0	-53.8	Total	S'	S''
-38.6	-3.4	-13.1	-55.1	+25.5	-44.9	D'	$D'.S'$	$D'.S''$
-55.0	+19.0	+2.9	-33.1	+57.9	-90.1	D''	$D''.S$	$D''.S''$

columns (1) the first line represents the totals of the three columns s_0, s_1, s_2 , of the d and s table (Table 46), the second line the differences $d_2 - d_0$ for each columns, and the third line the quantity $d_2 - 2d_1 + d_0$ for each column. Each number need only be written on the machine once, the sequence being:

$$\begin{array}{r}
 +217.8 \\
 -256.4 \\
 \hline
 -38.6 \\
 +256.4 \times 2 \\
 -264.6 \times 2 \\
 \hline
 -55.0 \\
 +264.6 \times 3 \\
 \hline
 738.8
 \end{array}$$

The computer must learn to read negative numbers directly from the machine.

A second application of the same process to the rows of (1) gives the required quantities (2) in the order shown.

11d. *General remarks.*

The method used in the above example is perfectly general, and can be used to subdivide the interactions in any manner corresponding to that adopted for the main effects. If the main effects are orthogonally divided then the interactions will also be orthogonally divided. Moreover there is no need to subdivide into single degrees of freedom. Thus if the factor a represents three varieties and b three levels of a fertilizer, we may subdivide into two pairs of degrees of freedom $A.B'$ and $A.B''$: the former will be given by the differences between the linear responses of the three varieties, the latter by the differences between the curvatures.

Subdivision of interaction degrees of freedom is useful, in the same manner as was the subdivision of main effects, for throwing into prominence effects which might otherwise escape notice. In the interaction of two fertilizers, for example, we should expect the component $A'.B'$ to be large compared with the remaining components, but if the four degrees of freedom are jointly tested its significance might be obscured. Subdivision is also useful when estimating the error from interactions, since we may reasonably expect interactions involving a component of curvature to be small even in cases where the component $A'.B'$ cannot legitimately be included in error. An example of this is provided by a single replication of a $3 \times 3 \times 3$ design.

12. THE $3 \times 3 \times 3$ DESIGN: SINGLE REPLICATION.

This particular design is of considerable practical importance in agricultural fertilizer trials, for it enables the optimal levels of all three standard fertilizers to be simultaneously investigated, and is not too large to be undertaken on ordinary commercial farms. We will therefore analyse the first replication of the sugar beet experiment already given, treating it as if it were the whole experiment.

12a. *Systematic method of analysis.*

Since experiments of this type are usually undertaken simultaneously at a number of centres, it is advisable to adhere to some systematic method of analysis and presentation of the results. In practice it has been found best in fertilizer trials to present the response to the double dressing of each factor (the linear response), the difference of the additional response to the second and the response to the first dressing (the curvature), and the linear component of interaction of each pair of factors, together with their standard errors.

The calculations proceed as follows:

1. Calculate the total sum of squares and the correction for the mean, obtaining the block totals at the same time.

2. Set out the yields as in Table 45, and calculate the five two-way tables similar to those of Table 46a, and thence the totals $[W]$, $[X]$, $[Y]$ and $[Z]$. The block totals check one of these sets, here $[Y]$. The table for d and s and the interaction totals are shown in Table 55.

TABLE 55. CALCULATION OF MAIN EFFECTS AND INTERACTIONS.

	50	51	52		W	X	Y	Z
d_0	144.0	145.2	142.8	432.0	402.6	403.5	439.9	420.3
d_1	143.3	139.1	129.1	411.5	396.5	415.3	425.8	397.4
d_2	128.1	137.5	115.6	381.2	425.6	405.9	359.0	407.0

3. Calculate the total sum of squares for each of the first three two-way tables, obtaining one set of marginal totals for each of these tables in the process, and also the sums of squares for [Y] (blocks) and for [W], [X] and [Z]. These sums of squares are shown in Table 56 (blocks in Table 58).

TABLE 56. AUXILIARY TABLE OF SUMS OF SQUARES.

D, S and $D.S$	262.05	S, N and $S.N$	250.82
D, N and $D.N$	333.49	W, X and Z	90.36

4. Calculate the totals for the linear responses and curvatures from the main-effect totals, and at the same time check the total of each set of main-effect totals. The method of Section 11c may be used. Thus $381.2 - 432.0 = -50.8$. $381.2 + 432.0 - 2 \times 411.5 = -9.8$, and the total (which need not be written down) $= 1224.7$. Enter the values obtained in Table 57. Then take the sum of squares of the linear response totals, dividing by 18, and the sum of squares of the curvature totals, dividing by 54, and enter in Table 58.

TABLE 57. MAIN EFFECTS AND INTERACTIONS.

Factor	Totals		cwt. per acre		Factors	Interactions	
	Linear response	Curvature	Linear response	Curvature		Total	cwt. per acre
D	-50.8	-9.8	-5.6	-1.1	$D'.S'$	-11.3	-1.9
S	-27.9	-40.7	-3.1	-4.5	$D'.N'$	-19.4	-3.2
N	+45.4	-32.0	+5.0	-3.6	$S'.N'$	+13.8	+2.3
St. error	± 14.4	± 25.0	± 1.6	± 2.8	St. error	± 11.8	± 2.0
Divisor	18	54			Divisor	12	

TABLE 58. ANALYSIS OF VARIANCE.

	D.F.	Sum of squares	Mean square
Blocks	2	415.03	207.52
Linear responses	3	301.12	
Curvatures	3	51.42	
Linear interactions	3	57.87	
Other interactions (error)	15	173.79	11.59
Total	26	999.23	

If the sum of squares for blocks and those of Table 56, less the sums of squares for the linear responses and curvatures, add up to the total sum of squares, the whole computation up to this point may be regarded as checked.

5. Calculate the totals for the linear components of the interactions from the cross differences of the corner values of the two-way tables, entering these in Table 57. Thus $144.0 + 115.6 - 128.1 - 142.8 = -11.3$. Divide the sum of squares by 12 and enter in Table 58. This calculation must be carefully checked.

6.. Calculate the error sum of squares by subtraction, and complete Table 58. Enter the standard errors of the totals in Table 57, e.g. $\sqrt{18 \times 11.59} = 14.4$. Then convert the values of Table 57 to the proper units. Here the conversion factor for the linear responses and the curvatures is $\frac{1}{6}$, and for the interactions is $\frac{1}{6}$, since the yields of the single plots are already in cwt. per acre.

This completes the analysis. Tests of significance can be made in the ordinary manner by the t test. The linear responses to change of sowing date and nitrogen are significant but that to spacing is barely so. The error mean square 11.59 agrees well with that already found from the analysis of the whole experiment.

12b. *Alternative method.*

An alternative method of analysis is to obtain all the main effects and two-factor interactions as single degrees of freedom by the procedure illustrated in Table 54. It will be noticed that each component of the main effects appears in two tables. The computation can therefore be slightly abbreviated by the omission of one set of main effects from each table. The total of each 3×3 table should, however, be checked.

If this procedure is adopted there is no need to compute the sums of squares for the 3×3 tables shown in Table 56. The final analysis of variance will appear in the form shown in Table 59, the whole computation being self-checking.

TABLE 59. ANALYSIS OF VARIANCE, ALTERNATIVE METHOD.

	D.F.	Sum of squares	Mean square
Blocks	2	415.03	207.52
Linear responses	3	301.12	
Curvatures	3	51.42	
Interactions: Linear \times linear ..	3	57.87	
Linear \times curv. ..	6	68.19	
Curv. \times curv. ..	3	15.22	
W, X, and Z ..	6	90.36	
		173.77	11.59
Total	26	999.23	

12c. *The linear component of the three-factor interaction.*

The linear component of the three-factor interaction in $3 \times 3 \times 3$ experiments has a certain interest, both because it is more likely to be of appreciable magnitude than any other component, and also because it represents the correcting term to the estimate of the combined responses to the highest levels of all three factors given by the sum of the three linear components of the main effects (Section 2e).*

*There are also other correcting terms in the $3 \times 3 \times 3$ system, but these are likely to be small.

At first sight its estimation in confounded experiments appears complicated. There is, however, no great difficulty, for we have the identity :

$$[A'.B'.C'] = \frac{1}{3} \{ [W_2] - [W_1] + [X_3] - [X_1] + [Y_3] - [Y_1] + [Z_3] - [Z_2] \},$$

as is easily verified from Table 43, or numerically from Tables 45 and 46b, ignoring the confounding.

In partially confounded experiments it is only necessary to substitute the corresponding totals, freed from confounding, which are denoted by dashes in Table 46, multiplying these by the necessary fraction to make them the equivalent of totals over the whole experiment. Thus in the example already given Y and Z are partially confounded and the totals $[Y']$ and $[Z']$ include only half the plots and must therefore be multiplied by 2. (If there were four replications, confounding all three-factor interactions, the multiplier would be $\frac{4}{3}$.) Thus in our example we have :

$$[D'.S'.N'] = \frac{1}{3} \{ 694.6 - 715.6 + 721.7 - 721.2 + 2(317.8 - 316.7) + 2(407.0 - 397.4) \} = + 0.3$$

and the error variance is

$$\frac{1}{3^2} \{ 18 + 18 + 18 + 18 + 2^2(9 + 9 + 9 + 9) \} \sigma^2 = 24\sigma^2.$$

Consequently, in units of a single plot yield, here cwt. per acre,

$$D'.S'.N' = \frac{1}{3} (+ 0.3) = + 0.04 \pm 2.24$$

since there are two replications, so that $[D'.S'.N']$ would be the difference of two sums of 8 plots each if there were no confounding. The same result may be reached (more laboriously) by using the table of adjusted yields (Section 10c).

If there were no confounding the error variance of $[A'.B'.C']$ would be $\frac{1}{6} (8 \times 18) \sigma^2 = 16\sigma^2$ so that $\frac{2}{3}$ of the relative information is retained. (With all components equally confounded $\frac{2}{4}$ of the relative information would be retained.)

If one set of components of the three-factor interaction is completely confounded, as must be the case in a single replication, clearly no estimate of the linear component is possible unless it is assumed that the remaining components are negligible. If this is assumed then each of the differences $[W_2] - [W_1]$, etc., provides a separate estimate of the linear component. Thus with a single replication only, when Y is confounded, as in the example just considered, we have

$$A'.B'.C' = \frac{1}{9} \{ [W_2] - [W_1] + [X_3] - [X_1] + [Z_3] - [Z_2] \},$$

the additional factor $\frac{4}{3}$ being introduced to compensate for the omission of one of the four estimates, together with a further factor $\frac{1}{4}$ to give $A'.B'.C'$ in terms of a single plot yield. Hence

$$D'.S'.N' = \frac{1}{9} \{ 396.5 - 402.6 + 405.9 - 403.5 + 407.0 - 397.4 \} \\ = \frac{1}{9} (+ 5.9) = + 0.66$$

The error variance of $A'.B'.C'$ is now given by

$$\frac{1}{81} (6 \times 9) \sigma^2 = \frac{2}{3} \sigma^2$$

so that the standard error of the estimate is here ± 2.78 . If there were no

confounding the error variance would be $\frac{1}{10} (8) \sigma^2 = \frac{1}{2} \sigma^2$. Consequently $\frac{3}{4}$ of the relative information is retained, but, of course, only on the assumption that the other components are negligible.

The sum of squares attributable to the corresponding single degree of freedom is given by

$$\frac{3}{2} \left[\frac{1}{8} (+ 5.9) \right]^2 = \frac{1}{24} (+ 5.9)^2 = 0.64$$

This can be deducted from the sum of squares for error in Table 58, leaving 14 degrees of freedom for error. Clearly in a series of experiments this deduction should be either made or not made consistently: it is not permissible to perform the deduction only when the error is reduced thereby.

The following alternative series of expressions (for a single replication, Y confounded) may be noted. If

$$Q = [A'.B'.C'] + \frac{1}{9}[Y_1] - \frac{1}{3}[Y_3]$$

or

$$3Q = 3[A'.B'.C'] + [Y_1] - [Y_3]$$

then

$$A'.B'.C' = \frac{1}{3}Q = \frac{1}{9}(3Q)$$

The error variance of this estimate is

$$\frac{2}{3}\sigma^2$$

and the sum of squares is

$$\frac{1}{3}Q^2 = \frac{1}{27}(3Q)^2$$

The above expressions are worth careful study. The total $[A'.B'.C']$, which would form the basis of the estimate in an unconfounded experiment, is corrected by the requisite fractions of the block totals $[Y_1]$ and $[Y_3]$ to eliminate block effects, giving Q . The fractional multipliers can then all be written down, if the relative information, here $\frac{3}{4}$, is known, by multiplying the fractions that would be used in an unconfounded experiment by the reciprocal of this relative information. Thus $\frac{1}{9} = \frac{4}{9} \times \frac{1}{4}$, $\frac{2}{9} = \frac{4}{9} \times \frac{1}{2}$ and $\frac{1}{3} = \frac{4}{9} \times \frac{1}{3}$. Note how $3Q$ is used in place of Q in the actual computation.

This method of adjustment by means of block totals forms the basis of the analytical methods applicable to confounded designs involving factors at both two and three levels, which are described in the following sections.

13. CONFOUNDING WITH SOME FACTORS AT TWO AND SOME AT THREE LEVELS.

Experiments containing factors at both two and three levels cannot be so simply confounded as those containing factors at two or at three levels only, because it is impossible to divide the treatment combinations into sets which correspond to the highest order interactions. The best designs are those which confine the confounding as much as possible to the highest order interactions. These designs necessarily involve the partial confounding of the more important interactions also, the confounded degree or degrees of freedom in any replication being divided between different sets of treatment degrees of freedom. The fraction of the information sacrificed on the more important interaction is, however, quite small.

Designs of this type are not quite so simple to analyse as designs of the 2^n or 3^n types. The designs must be balanced, and therefore the number of replications used must be some multiple of the number required for a balanced arrangement. The computation is similar for all the different patterns. An example is given for the $3 \times 2 \times 2$ design, which will illustrate the use of the formulæ.

13a. Statistical analysis of $3 \times 2 \times 2$ design.

Denote the three factors by $A(0, 1, 2)$, $B(0, 1)$, $C(0, 1)$. Since 4 is not a factor of 6 it is clear that the interaction $B.C$ cannot be completely unconfounded if the experiment is arranged in blocks of 6 plots. The design of Table 60 confounds $B.C$ as little as possible.

TABLE 60. $3 \times 2 \times 2$ DESIGN IN BLOCKS OF 6 PLOTS.

Ia			Ib			IIa			IIb			IIIa			IIIb		
a	b	c	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
0	0	1	0	0	■	0	0	0	0	■	1	0	0	0	0	0	1
■	1	0	0	1	1	0	1	1	0	1	0	0	1	1	0	1	0
1	0	0	1	0	1	1	0	1	1	0	0	1	0	0	1	0	1
1	1	1	1	1	■	1	1	0	1	1	1	1	1	1	1	1	0
2	0	0	2	0	1	2	0	0	2	0	1	2	0	1	2	0	0
2	1	1	2	1	0	2	1	1	2	1	0	2	1	0	2	1	1

The interactions $B.C$ and $A.B.C$ are partially confounded with block differences in each replication, since the actual degree of freedom confounded lacks orthogonality with both these sets. In each replication the confounding is different, the three replications giving a balanced design which enables the treatment degrees of freedom $B.C$ and $A.B.C$ to be estimated without difficulty.

13b. Statistical analysis of $3 \times 2 \times 2$ design.

Since the interaction $B.C$ is partially confounded it is necessary to correct the ordinary interaction total $[B.C]$ by means of the block totals $[Ia]$, $[Ib]$, etc. If

$$[Ib] - [Ia] = g_1, [IIb] - [IIa] = g_2, [IIIb] - [IIIa] = g_3,$$

and if we calculate

$$3Q = 3[B.C] + g_1 + g_2 + g_3$$

it can easily be verified that Q is unaffected by block differences or treatment effects other than $B.C$.

The estimate of $B.C$ in units of the yield of a single plot is given by

$$B.C = \frac{1}{16} Q = \frac{1}{48} (3Q)$$

when there are 36 plots. The error variance of $B.C$ is $\frac{1}{8}\sigma^2$. Note that in an unconfounded experiment the estimate and error variance would be $\frac{1}{16} [B.C]$ and $\frac{1}{8}\sigma^2$. The corresponding sum of squares is

$$\frac{1}{32} Q^2 = \frac{1}{288} (3Q)^2$$

as compared with $\frac{1}{36} [B.C]^2$ in an unconfounded experiment. The relative information is given by the ratio

$$\frac{1/8}{1/36} = \frac{9}{8}$$

Thus $\frac{1}{3}$ of the information is lost by the confounding when there is no reduction in the error variance per plot.

The estimate of $A.B.C$ is obtained in a similar manner. Calculate the three quantities

$$\begin{aligned} 3R_0 &= 3 [B.C.a_0] - g_1 + g_2 + g_3 \\ 3R_1 &= 3 [B.C.a_1] + g_1 - g_2 + g_3 \\ 3R_2 &= 3 [B.C.a_2] + g_1 + g_2 - g_3 \end{aligned}$$

with the check that $3R_0 + 3R_1 + 3R_2 = 3Q$.

The interaction $A.B.C$, in units of a single plot yield, is given by

$$A.B.C = \frac{1}{10} \text{dev} (R_0, R_1, R_2) = \frac{1}{10} \text{dev} (3R_0, 3R_1, 3R_2)$$

as compared with $\frac{1}{6} [B.C.a_0]$, etc., in an unconfounded experiment. The error variance applicable to each of these quantities is $\frac{3}{5}\sigma^2$, as compared with $\frac{1}{3}\sigma^2$. The sum of squares is given by

$$\frac{3}{5} \text{dev}^2 (R_0, R_1, R_2) = \frac{1}{5} \text{dev}^2 (3R_0, 3R_1, 3R_2)$$

The relative information is given by the ratio

$$\frac{1/3}{3/5} = \frac{5}{9}$$

and the relative loss of information on each of the two degrees of freedom is therefore $\frac{4}{9}$. Note that

$$1 \times \frac{1}{3} + 2 \times \frac{4}{9} = 1$$

corresponding to the single degree of freedom confounded in each replication. This is a property of balanced arrangements.

The reader will find it instructive to construct the above formulæ by means of the rule given at the end of the last section, using only the fractions representing the relative information.

13c. Example.

The plan and yields of the experiment on potatoes already referred to in Section 9d (1/65 acre plots) are given in Table 61.

TABLE 61. PLAN AND YIELDS (LB.) OF $3 \times 2 \times 2$ EXPERIMENT.

Ia		Ib		IIa	
n_a 172	n_{op} 161	n_{1p} 231	n_o 166	n_{omp} 208	n_{amp} 144
n_{om} 192	n_1 145	n_{ap} 204	n_{omp} 253	n_{1m} 190	n_a 104
n_{amp} 227	n_{1mp} 232	n_{1m} 231	n_{am} 214	n_o 113	n_{1p} 131
n_1 176	n_{am} 186	n_{1m} 238	n_{om} 198	n_1 158	n_{1mp} 171
n_o 132	n_{1mp} 242	n_{op} 180	n_a 175	n_{om} 171	n_{ap} 135
n_{omp} 196	n_{ap} 178	n_{amp} 230	n_{1p} 216	n_{am} 146	n_{op} 103
IIIa		IIIb		IIb	

Using the results already obtained in Table 38 we have

$$3Q = 3(-61) + 170 - 6 + 127 = +108$$

$$3R_0 = 3(+63) - 170 - 6 + 127 = +140$$

$$3R_1 = 3(-113) + 170 + 6 + 127 = -36$$

$$3R_2 = 3(-11) + 170 - 6 - 127 = +4$$

$$P.M = +2.25 = +0.07 \text{ tons per acre.}$$

$$P.M.N = +10.4, -7.2, -3.2 = +0.30, -0.20, -0.10 \text{ tons per acre.}$$

The sums of squares are :

	D.F.	Sum of squares	Mean square
<i>P.M</i>	1	40.5	40.5
<i>P.M.N</i>	2	283.7	141.9

Replacing the values already given in Table 39 by these, we can complete the analysis as shown in Table 62.

TABLE 62. ANALYSIS OF VARIANCE OF $3 \times 2 \times 2$ EXPERIMENT.

				D.F.	Sum of squares	Mean square
Blocks	5	24938.9	2017.0
Treatments	11	26302.1	2391.1
Error	19	6363.8	334.9
Total	35	57604.8	

Two two-way tables will be required to show the interactions between p and n and m and n . Since these interactions are not affected by the confounding the tables can be obtained directly from Table 38 in the manner already described. If a two-way table for p and m is also required it can best be built up by the method of Section 3, using the value of $P.M$ calculated above. These three tables are given in Table 63.

TABLE 63. TWO-FACTOR TABLES, TONS PER ACRE.

	n_0	n_1	n_2		(1)	p
(1)	4.14	5.11	4.68	4.64	4.41	4.88
m	5.89	6.31	5.55	5.92	5.61	6.22
	5.01	5.71	5.12	5.28	5.01	5.55
(1)	4.70	5.50	4.82	5.01		
p	5.32	5.91	5.41	5.55		

Since no one of the interactions between two factors is significant it will scarcely be necessary to give a three-way table to exhibit the interactions between all three factors, but if one is required the calculation may be carried out in two stages as in the $3 \times 3 \times 3$ example (Section 10c). Thus, neglecting the interaction $N.M.P$,

$$n_0mp = 5.89 + 5.32 + 6.22 - 5.01 - 5.92 - 5.55 + 5.28 = 6.23$$

To include the effect of $N.M.P$ one half the values already obtained for this interaction must be added to the lines (1) and mp and subtracted from the lines p and m , thus

$$n_0mp = 6.23 + 0.15 = 6.38$$

The full sets of values are shown in Table 64.

TABLE 64. THREE-FACTOR TABLE, TONS PER ACRE.

(a) Neglecting <i>N.M.P</i>				(b) Including <i>N.M.P</i>			
	<i>n</i> ₀	<i>n</i> ₁	<i>n</i> ₂		<i>n</i> ₀	<i>n</i> ₁	<i>n</i> ₂
(1)	3.87	4.94	4.42	(1)	4.02	4.84	4.37
<i>p</i>	4.42	5.28	4.94	<i>p</i>	4.27	5.38	4.99
<i>m</i>	5.54	6.06	5.21	<i>m</i>	5.39	6.16	5.26
<i>mp</i>	6.23	6.54	5.87	<i>mp</i>	6.38	6.44	5.82

13d. $3 \times 2 \times 2 \times 2$ design in blocks of 6 plots.

With three factors at two levels (but not with more) there is an arrangement in blocks of 6 plots similar to that with two factors at two levels, only $\frac{1}{6}$ of the relative information on the interactions between pairs of factors at two levels being sacrificed. 72 plots are required to provide a balanced design. The 12 blocks of this design are given in Table 65.

TABLE 65. $3 \times 2 \times 2 \times 2$ DESIGN.

Level of <i>a</i>	Ia	Ib	Ic	Id	IIa	IIb	IIc	IId	IIIa	IIIb	IIIc	IIId
<i>a</i> ₀	<i>b</i>	(1)	<i>d</i>	<i>c</i>	<i>c</i>	<i>d</i>	(1)	<i>b</i>	<i>d</i>	<i>c</i>	<i>b</i>	(1)
<i>a</i> ₁	<i>c</i>	<i>d</i>	(1)	<i>b</i>	<i>d</i>	<i>c</i>	<i>b</i>	(1)	<i>b</i>	(1)	<i>d</i>	<i>c</i>
<i>a</i> ₂	<i>d</i>	<i>c</i>	<i>b</i>	(1)	<i>b</i>	(1)	<i>d</i>	<i>c</i>	<i>c</i>	<i>d</i>	(1)	<i>b</i>

In this table only one of the pair of combinations of *b*, *c* and *d* for each level of *a* is shown. When (1) occurs *bcd* must occur also; similarly *cd* must occur with *b*, *bd* with *c* and *bc* with *d*. Thus the block Ib contains the treatments *a*₀, *a*₀*bcd*, *a*₁*d*, *a*₁*bc*, *a*₂*c*, *a*₂*bd*.

The required formulæ are simple extensions of those applicable to the $3 \times 2 \times 2$ design. Denote the differences between the block totals in replication I by *g*₁, *g*₁' and *g*₁'', where

$$\begin{aligned} g_1 &= [Ia] + [Ib] - [Ic] - [Id] \\ g_1' &= [Ia] - [Ib] + [Ic] - [Id] \\ g_1'' &= [Ia] - [Ib] - [Ic] + [Id] \end{aligned}$$

with similar expressions for replications II and III.

To estimate the interactions *C.D*, *B.D* and *B.C*, the quantity *Q* must be replaced in turn by

$$\begin{aligned} 3Q &= 3[C.D] + g_1 + g_2 + g_3 \\ 3Q' &= 3[B.D] + g_1' + g_2' + g_3' \\ 3Q'' &= 3[B.C] + g_1'' + g_2'' + g_3'' \end{aligned}$$

The three-factor interactions are obtained in the same way, the formulæ being identical with those already given except for the introduction of dashes.

The remainder of the computation proceeds as before, except that all divisors must be doubled to allow for the increase in the number of plots.

13e. Extension to 3×2^n in blocks of $3 \times 2^{n-1}$ and $3 \times 2^{n-2}$.

With blocks of $3 \times 2^{n-1}$ the methods and equations set out for the $3 \times 2 \times 2$ design in blocks of 6 plots are immediately applicable. Take *X*₁ to represent

the treatment combinations which are taken as positive in the interaction between the n factors at two levels, and X_0 the combinations which are taken as negative, so that with three factors b , c , and d at two levels, X_1 represents the four combinations bcd , b , c , d , and X_0 the four combinations bc , bd , cd , (1). As before three complete replications are necessary, the six blocks being those shown in Table 66.

TABLE 66. 3×2^n DESIGN IN BLOCKS OF $3 \times 2^{n-1}$ PLOTS.

Ia	Ib	IIa	IIb	IIIa	IIIb
a_0X_0	a_0X_1	a_0X_1	a_0X_0	a_0X_1	a_0X_0
a_1X_1	a_1X_0	a_1X_0	a_1X_1	a_1X_1	a_1X_0
a_2X_1	a_2X_0	a_2X_1	a_2X_0	a_2X_0	a_2X_1

The interaction between all the factors at two levels and the interaction between these and the factor at three levels, will be partially confounded. The only modification required in the formulæ already given is a proportionate increase in the numerical divisors to allow for the increased number of plots.

The extension of the $3 \times 2 \times 2 \times 2$ design follows exactly the same lines as the extension of the $3 \times 2 \times 2$ design, giving blocks of $3 \times 2^{n-2}$. If, for example, a fourth factor e at two levels is introduced the interactions $B.C.E$, $B.D.E$ and $C.D$, and their interactions with A , might be chosen for partial confounding. The design is given by writing b and e for b , and (1) and be for no b , in the $3 \times 2 \times 2 \times 2$ design. Thus block Ia will contain the plots

a_0b , a_0e , a_0cd , a_0bcde , a_1c , a_1bce , a_1bd , a_1de , a_2d , a_2bde , a_2bc , a_2ce .

It may be noted that there is no $3 \times 2 \times 2$ design in a 6×6 quasi-Latin square which leaves the main effects completely unconfounded. A design exists which partially confounds the interaction between the two factors at two levels and the interactions between all three factors, and in addition slightly confounds the main effect of one of the factors at two levels. In view of the additional complication in the computations we have omitted this design.

13f. $3 \times 3 \times 2$ design.

Denote the three factors by A (0, 1, 2), B (0, 1, 2), C (0, 1). Since 9 is not a factor of 6 the interaction $A.B$ cannot be completely unconfounded when the experiment is arranged in blocks of 6 plots. Using I and J to indicate the different diagonal sets of the combinations of a and b , as indicated in Table 40, we have the following design of 36 plots (Table 67) which partially confounds $A.B$ (I) and $A.B.C$ (I).

TABLE 67. $3 \times 3 \times 2$ DESIGN IN BLOCKS OF 6 PLOTS.

	Ia	Ib	Ic	IIa	IIb	IIc
c_0	I_2	I_3	I_1	I_3	I_1	I_2
c_1	I_3	I_1	I_2	I_2	I_3	I_1

The first block, for example, will contain the treatments

$$a_1 b_0 c_0, a_2 b_1 c_0, a_0 b_2 c_0, a_2 b_0 c_1, a_0 b_1 c_1, a_1 b_2 c_1.$$

A similar design, which confounds $A.B$ (\mathcal{J}) and $A.B.C$ (\mathcal{J}), is obtained by writing \mathcal{J} instead of I . If 72 plots are available both designs should be used, so that all four degrees of freedom for $A.B$, and also all four for $A.B.C$, are equally confounded.

The method of analysis is similar to that applicable to the $3 \times 2 \times 2$ design. To estimate the I component of $A.B$ when there are 36 plots and the I components are confounded the quantity

$$2Q_1 = 2[I_1] - [Ib] - [Ic] - [IIb] - [IIc]$$

and two similar quantities $2Q_2$ and $2Q_3$ may be calculated. The sum of these is zero.

The relative information is $\frac{2}{3}$, so that, since $\frac{1}{3} \times \frac{1}{2} = \frac{1}{6}$, the estimate of the interactions is given by

$$A.B(I) = \frac{1}{6}(Q_1, Q_2, Q_3) = \frac{1}{18}(2Q_1, 2Q_2, 2Q_3)$$

the error variance of each of these quantities being $\frac{1}{3} \sigma^2$. The sum of squares for the two degrees of freedom is

$$\frac{1}{6} S(Q^2) = \frac{1}{36} S(2Q)^2$$

The estimates of the confounded components of $A.B.C$ are obtained by calculating the three quantities

$$2R_1 = 2[I_1.C] - [Ib] + [Ic] + [IIb] - [IIc]$$

etc., where $[I_1.C]$ denotes the sum of the I_1 components in the table of $c_1 - c_0$. The sum of the three quantities is $2[C]$. The relative information is $\frac{1}{4}$, so that, since $\frac{1}{4} \times \frac{1}{6} = \frac{1}{24}$, the estimate is given by

$$A.B.C(I) = \frac{2}{3} \text{dev } R = \frac{1}{3} \text{dev } 2R$$

Note the introduction of an extra 2, since one of the factors is at two levels only.

The error variance of each of these quantities is $\frac{1}{4} \times \frac{1}{3} \sigma^2 = \frac{1}{12} \sigma^2$.

The sum of squares for the two degrees of freedom is

$$\frac{1}{3} \text{dev}^2 R = \frac{1}{12} \text{dev}^2 2R$$

The formulæ for the design of 36 plots which confounds the \mathcal{J} components of interaction are obtained from the above formulæ by writing \mathcal{J} for I .

If 72 plots are available and both the I and \mathcal{J} components are confounded, then to estimate $A.B(I)$ the quantities Q are calculated as above, but each total $[I]$ is taken over the whole experiment and therefore includes 24 plots. The relative information is now $\frac{1}{6}$, so that the divisor 9 in the above formulæ must be replaced by 21. Estimates for $A.B(\mathcal{J})$ are similarly obtained. Estimates for $A.B.C(I)$ are obtained by calculating quantities R as above, but the relative information is now $\frac{1}{8}$, so that all the divisors given above must be multiplied by 5.

13g. $3 \times 3 \times 3 \times 2$ design in blocks of 6 plots.

There are four designs, each of 108 plots (two replications), in which the interactions of all pairs of factors at three levels are partially confounded in the same manner as in the $3 \times 3 \times 2$ design in 36 plots. In each of the designs two degrees of freedom of the interaction between all three factors at three levels are completely confounded. The actual sets of confounded degrees of freedom are given in Table 68.

TABLE 68. CONFOUNDED DEGREES OF FREEDOM IN $3 \times 3 \times 3 \times 2$ DESIGNS.

	Design :	"W"	"X"	"Y"	"Z"
Partially confounded	$\begin{cases} A.B \text{ and } A.B.D \\ A.C \text{ and } A.C.D \\ B.C \text{ and } B.C.D \end{cases}$	$\begin{matrix} \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \end{matrix}$	$\begin{matrix} \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \end{matrix}$	$\begin{matrix} \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \end{matrix}$	$\begin{matrix} \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \\ \cdot\cdot & \cdot\cdot & \cdot\cdot & \cdot\cdot \end{matrix}$
Completely confounded	$A.B.C$	W	X	Y	Z

From this table it will be seen that if all four designs are used (432 plots) the I and J components of all the partially confounded interactions are confounded equally, so that, as in the $3 \times 3 \times 2$ design in 72 plots, the relative information on $A.B$, $A.C$ and $A.D$ is $\frac{7}{8}$, and that on $A.B.D$, $A.C.D$ and $B.C.D$ is $\frac{5}{8}$. In addition all components of $A.B.C$ are equally confounded, the relative information being $\frac{3}{4}$.

If the 27 combinations of the three-level factors are divided into the following 9 sets of three:

K_1	K_2	K_3	K_4	K_5	K_6	K_7	K_8	K_9
000	100	200	010	110	210	020	120	220
111	211	011	121	221	021	101	201	001
222	022	122	202	002	102	212	012	112

then the first 9 blocks of the "Z" design are those given in Table 69, the other 9 blocks being obtained by interchanging d_0 and d_1 .

TABLE 69. FIRST REPLICATION OF THE $3 \times 3 \times 3 \times 2$ "Z" DESIGN.

Block	Ia	Ib	Ic	Id	Ie	If	Ig	Ih	Ii
d_0	K_1	K_6	K_8	K_2	K_4	K_9	K_3	K_5	K_7
d_1	K_6	K_8	K_1	K_4	K_9	K_2	K_5	K_7	K_3

The "W," "X" and "Y" designs are obtained from the "Z" design by interchanging a_1 and a_2 , b_1 and b_2 , and c_1 and c_2 respectively in the expression for the K 's. Thus for "W" we take K_1 to represent the combination 000, 211 and 122, etc.

The estimates of the partially confounded effects are obtained in exactly the same manner as in the $3 \times 3 \times 2$ design. Thus to estimate $A.B$ (I) in the "Z" design the quantity

$$2 Q_1 = 2 [I_1]_{A.B} - [Ia] - [Ic] - [Ie] - [If] - [Ig] - [Ih] - [IIa] - [IIc] - [IIe] - [IIf] - [IIg] - [IIh]$$

and two similar quantities are calculated.

13h. Extension to $3^n \times 2$ designs in blocks of $3^{n-1} \times 2$ and $3^{n-2} \times 2$ plots.

The designs already given can be extended in the same manner as the $3 \times 3 \times 3$ and $3 \times 3 \times 3 \times 3$ designs (Section 10f).

It may be noted here that there is no reasonably simple $3 \times 3 \times 2 \times 2$ design in blocks of 6 plots. A design in blocks of 12 plots (and more generally a design for $3 \times 3 \times 2^n$ in blocks of $3 \times 3 \times 2^{n-1}$ plots) may be obtained by extending the $3 \times 3 \times 2$ design in the same manner as the extension of the $3 \times 2 \times 2$ design to 3×2^n in blocks of $3 \times 2^{n-1}$. This design confounds $A.B$ and $A.B.C.D$

only, but there are other designs which sacrifice less information on $A.B$, at the expense of confounding $A.B.C$ and $A.B.D$, and generally increasing the complications of the computations. We shall not consider them here.

13i. $3 \times 3 \times 2$ design in a 6×6 quasi-Latin square.

It is possible to form a square of which the rows are the blocks of Table 67, and thus confound the I components of the interactions between the two factors at three levels, and the columns are the similar blocks which confound the J components of these interactions. Only one such square exists (except for permutations of rows and columns). This square is shown in Table 70, where the first figure of each number indicates the combination of the two three-level factors, and the second figure the level of the factor at two levels.

TABLE 70. $3 \times 3 \times 2$ DESIGN IN A 6×6 QUASI-LATIN SQUARE.

20	70	60	41	31	81
40	30	80	91	51	11
90	50	10	21	71	61
71	61	21	30	80	40
31	81	41	50	10	90
51	11	91	70	60	20

The estimates of the confounded interactions are computed in exactly the same manner as in the $3 \times 3 \times 2$ design in blocks of 6 plots, using row and column totals instead of block totals. The relative information on the interactions between the two three-level factors is $\frac{3}{4}$, and that on the interactions of all three factors is $\frac{1}{4}$.

In this design there are only 8 degrees of freedom for error, but in view of the small amount of information available on the three-factor interactions these may justifiably be included in the estimate of error, giving 12 degrees of freedom in all, except in cases in which these interactions are likely to be large. This saves an appreciable amount of computation.

14. CONFOUNDING WITH ONE OR MORE FACTORS AT FOUR LEVELS OR EIGHT LEVELS.

14a. General method.

Since 4 and 8 are powers of 2 the possible systems of confounding when one or more factors are at four or eight levels and the remainder are at two levels can be derived quite simply by the general rule already given for factors at two levels only.

With any factor a at four levels there are associated three degrees of freedom, which may be partitioned into single degrees of freedom as follows:

$$A' = a_3 + a_2 - a_1 - a_0$$

$$A'' = a_3 - a_2 - a_1 + a_0$$

$$A''' = a_3 - a_2 + a_1 - a_0$$

The dashes are here used in a slightly different sense from those in Section 11. A'' represents the quadratic component of regression, $2A' + A'''$ represents the linear component, and $2A''' - A'$ represents the cubic component. If A''' is

confounded and the cubic component is assumed to be negligible then $\frac{1}{4}A'$ gives an estimate of the linear component of the regression.

Using this partition, A' and A'' may be taken as representing the main effects of two different two-level factors, in which case A''' will be their interaction. A single factor at four levels may thus be formally replaced by two factors at two levels. In a similar manner, a factor at eight levels may be replaced by three factors at two levels.

14b. Example: 4×4 designs.

As an example we may consider the design of a 4×4 experiment (factors a and b) in blocks of 8 and in blocks of 4 plots.

A 4×4 design is the equivalent of a 2^4 design. With blocks of 8 plots any single degree of freedom for interactions between the four two-level factors may be confounded. We might, for instance, confound $A'.A''.B'.B''$, which is equivalent to $A'''.B'''$. This would be the best single degree of freedom to choose if we wished to keep the linear and quadratic components of interaction as free as possible, without resorting to partial confounding. The partition of the treatment combinations into the two types of sub-block would then be given by the + and - signs in the product:

$$A'''.B''' = (a_3 - a_2 + a_1 - a_0) (b_3 - b_2 + b_1 - b_0).$$

A better course, however, would be to confound different interactions in different blocks. If four replications were available, for example, we might confound $A''.B''$, $A''.B'''$, $A'''.B''$ and $A'''.B'''$ once each.

With blocks of four plots three degrees of freedom will be confounded in each replication. With three replications the nine degrees of freedom representing interactions between A and B may be confounded in three sets. One such group of sets is:

$A'.B'$	$A'.B''$	$A'.B'''$
$A''.B''$	$A''.B'''$	$A''.B'$
$A'''.B'''$	$A'''.B'$	$A'''.B''$

The partition of the treatment combinations corresponding to the first set, for instance, is given by the four combinations of + and - signs, ++, +-, -+, --, in the two products $A'.B'$ and $A''.B''$. The three sets correspond to an orthogonal set of 4×4 Latin squares, with the rows and columns representing the four levels of the factors a and b respectively.

A balanced arrangement of this type is particularly useful when one of the factors represents four different varieties, or other treatments for which all possible comparisons are of equal interest, for in such a case the interactions of A' , A'' and A''' with B are all of equal importance.

14c. Combined varietal and manuring trials in Latin squares.

There is not space here to give a complete enumeration of designs including all the various combinations of factors at 2, 4 and 8 levels, but with the above example in mind the reader should have no difficulty in constructing the design he requires from the designs for factors at two levels given in Sections 5 and 8.

In particular he should notice the possibilities of arranging combined varietal and manuring (or cultivation) trials in 8×8 Latin squares.

Thus, for example, if four varieties and three fertilizers n, p, k are to be tested, the design of Table 32 may be used, identifying the combinations of b and c with the varieties, and a, d and e with n, p and k respectively. If V' , etc. are defined as A' etc. above, and V' is identified with B , and V'' with C , so that the combinations (1), b, c, bc of b and c are replaced by v_1, v_2, v_3, v_4 respectively, the following degrees of freedom will be confounded:

Rows: $V'.N.K, V''.P.K, V'''.N.P$
Columns: $V'.P.K, V''.N.P, V'''.N.K$

With 8 varieties and the three standard fertilizers, variants of the design of Table 33 may be used. If the combinations of a, b and c are identified with the varieties, and d, e and f with n, p and k respectively, we shall then have the following degrees of freedom confounded:

Rows: $V^1.N.K, V^2.N.P, V^3.P.K, V^4.N.P.K, V^5.P, V^6.K, V^7.N$
Columns: $V^1.N.P, V^2.N.P.K, V^3.K, V^4.P.K, V^5.N.K, V^6.N, V^7.P$

V^1, V^2, \dots, V^7 being a set of 7 orthogonal varietal degrees of freedom of the form

$$V^1 = v_1 - v_2 + v_3 - v_4 + v_5 - v_6 + v_7 - v_8$$

etc., such that $V^1.V^2 = V^3$, etc. A second square can be formed by making the cyclical change of varieties:

$$1 \rightarrow 3 \rightarrow 4 \rightarrow 5 \rightarrow 2 \rightarrow 6 \rightarrow 7 \rightarrow 1,$$

8 being left unchanged. The square so formed will confound an entirely different set of interactions. A further application of the same cyclical change will confound a further set different from the first two, but complete balance will only be obtained by using all the seven squares given by repetition of the above cyclical change, when each interaction degree of freedom between manures and varieties will be confounded twice, $\frac{5}{7}$ of the relative information being thus retained.

The reader who is interested in the structure of these designs will do well to determine their connection with an orthogonal set of seven 8×8 squares, such as that given in *The Design of Experiments* (2nd edition), or in *Statistical Tables for Biological, Medical and Agricultural Research*. He may note further that the pair of squares proposed in Section 8c for the design in 128 plots is not derivable from four squares of an orthogonal set, and should satisfy himself as to the reasons for this.

There is a set of similar designs for 9 varieties and 9 treatment combinations in 9×9 squares. One such square is that given in Table 51, the first number of each pair being now taken to represent the variety. By performing in turn on the *original* square the following interchanges:

- (1) 2 and 3, 4 and 7, 5 and 9, 6 and 8,
- (2) 2 \rightarrow 6 \rightarrow 3 \rightarrow 8 \rightarrow 2, 4 \rightarrow 5 \rightarrow 7 \rightarrow 9 \rightarrow 4,
- (3) 2 \rightarrow 8 \rightarrow 3 \rightarrow 6 \rightarrow 2, 4 \rightarrow 9 \rightarrow 7 \rightarrow 5 \rightarrow 4,

we generate three new squares. Balance is attained, for the four squares will between them equally confound all components of interaction between treatments and varieties, $\frac{3}{4}$ of the relative information being retained.

15. DUMMY TREATMENTS.

It frequently happens in factorial experiments that one or more of the factors is of such a nature that certain treatment combinations are identical. Thus if one of the factors consists of three different qualities of a fertilizer and another consists of three different amounts of the same fertilizer (including no fertilizer), there will in fact be no difference between the different qualities at zero level of the fertilizer. If the formal factorial design is followed, three identical plots having no fertilizer will be included in each replication. There will consequently be additional degrees of freedom for error arising from comparisons between identical combinations, and correspondingly fewer treatment degrees of freedom. The partition of the treatment degrees of freedom into their separate components will also be different. Confounding, moreover, introduces further complication.

There is not space here to discuss all the modifications that are required in the analysis of variance, if this analysis be conducted on strictly rigorous lines, but we will give certain arrangements of this type which will illustrate the main points.

Possible types of confounding are derivable from the ordinary factorial designs already given, by using dummy treatments where necessary. Other types not so derivable may also occasionally be of interest. For an example of these latter see (8).

15a. Application of fertilizer at two different times.

As a first example let us consider the design of an experiment to determine the response of sugar-beet to nitrogen applied at two different times, in conjunction with early and late lifting of the crop.

A $2 \times 2 \times 2$ design, with factors n , time of application, and time of lifting, might be adopted. This would give the treatment combinations

$$e, e', l, l', en, en', ln, ln'$$

where the dash indicates the later application of n , and e and l indicate early and late lifting. The combinations e and e' , and l and l' , are in reality identical.

It is not difficult to see that the appropriate partition of the degrees of freedom, and the estimates of the corresponding effects, are those given in Table 71.

TABLE 71. PARTITION OF DEGREES OF FREEDOM.

Effect					Estimate
Nitrogen (N)	$\frac{1}{4}(en + en' + ln + ln' - e - e' - l - l')$
Time of application (A)	$\frac{1}{2}(en - en' + ln - ln')$
Time of lifting (L)	$\frac{1}{4}(en + en' - ln - ln' + e + e' - l - l')$
$N.L$	$\frac{1}{4}(en + en' - ln - ln' - e - e' + l + l')$
$A.L$	$\frac{1}{2}(en - en' - ln + ln')$

These degrees of freedom are all orthogonal, and the sums of squares, plus the sums of square from $e - e'$ and $l - l'$, which are components of error, will total to the sum of squares for the seven degrees of freedom obtained from the treatment totals by keeping e and e' and l and l' separate.

If the experiment is arranged in blocks of 4 plots the confounding of the formal three-factor interaction will give the two block types

$$\begin{array}{cc} e & e' \\ l' & l \\ en' & en \\ ln & ln' \end{array}$$

The expression for $A.L$ above is now not orthogonal with blocks. It may be replaced by the formal expression for the $A.L$ interaction (with the numerical factor changed), namely

$$\frac{1}{2}(en - en' - ln + ln' + e - e' - l + l')$$

which is orthogonal with blocks. The function of the plots without n is to act as compensators for any inequalities between blocks. It is clear that with the same error variance per plot the variance of the estimate of this interaction will be doubled by the confounding.

There is now one error degree of freedom

$$e - e' + l - l'$$

the other being absorbed by the confounding. The reader will do well to set out the formal expressions derived from the ordinary $2 \times 2 \times 2$ design for all the degrees of freedom. He will find that the above error degree of freedom is twice the difference of the formal expressions for A and $N.A$, while the estimate of A in Table 71 is the sum of these expressions.

15b. Alternative designs.

It is instructive also to consider alternative designs for the above experiment. If the main interest of the experiment is a comparison of the effects of early and late application of nitrogen the above design may be considered unsuitable in that only one half of the plots contribute information on this point. An alternative set of treatments would be

$$e, l, en, en', ln, ln'$$

one of each of the duplicates being omitted.

The estimates of the treatment effects will then be those given in Table 72.

TABLE 72. PARTITION OF DEGREES OF FREEDOM.

Effect		Estimate
N	$\frac{1}{4}(en + en' + ln + ln' - 2e - 2l)$
A	$\frac{1}{2}(en - en' + ln - ln')$
L	$\frac{1}{2}(en + en' - ln - ln' + e - l)$
$N.L$	$\frac{1}{4}(en + en' - ln - ln' - 2e + 2l)$
$A.L$	$\frac{1}{2}(en - en' - ln + ln')$

These estimates are orthogonal. Note, however, that if N and L interact, L as here defined will be different from the L in the previous design.

Another design including the same treatments is that given by the $2 \times 2 \times 2$ design containing factors n early, n late, and time of lifting. The treatment combinations will then be

$$e, l, en, ln, en', ln', enn', lnn'$$

Here again only half the plots enter into the comparisons on time of application, but one quarter of the plots receive a double dressing of nitrogen, thus giving an estimate of the curvature of the response curve. The appropriate partition of the degrees of freedom is given in Table 73.

TABLE 73. PARTITION OF DEGREES OF FREEDOM.

Effect	Estimate
Response to double dressing (N') ..	$\frac{1}{2}(enn' + lnn' - e - l)$
Curvature (N'')	$\frac{1}{2}(enn' + lnn' - en - ln - en' - ln' + e + l)$
Time of application (A)	$\frac{1}{2}(en + ln - en' - ln')$
Time of lifting (L)	$\frac{1}{2}(enn' - lnn' + en - ln + en' - ln' + e - l)$
$L.N'$	$\frac{1}{2}(enn' - lnn' - e + l)$
$L.N''$	$\frac{1}{2}(enn' - lnn' - en + ln - en' + ln' + e - l)$
$L.A$	$\frac{1}{2}(en - ln - en' + ln')$

If the formal three-factor interaction is confounded this is equivalent to confounding $L.N''$. If the formal two-factor interactions between time of lifting and n early, and time of lifting and n late, are also confounded in their turn, each of the three equally frequently, two-thirds the relative information on $L.N'$, $L.N''$ and $L.A$ will be obtained. The above two-factor interactions are, in fact, $\frac{1}{2}(L.N' + L.A)$ and $\frac{1}{2}(L.N' - L.A)$.

15c. $3 \times 3 \times 3$ design including quality differences.

If we wish to experiment on three forms of nitrogen, each form being at three levels, in conjunction with three levels of phosphate, the ordinary $3 \times 3 \times 3$ design will give three sets of three identical treatment combinations.

The partition of the treatment degrees of freedom (including dummies) will therefore be as follows:

N	2	Q	2	$Q.N.P$	4
P	2	$Q.N$	2	Error	6
$N.P$	4	$Q.P$	4		

N , P and $N.P$ are estimated in the ordinary manner from the 3×3 table for n and p . Q and $Q.N$ will be estimated from the 3×2 table for q_0, q_1 and q_2 , and n_1 and n_2 (n_0 being omitted).

It may be reasonable to suppose that the differences due to quality at the higher level of n are double those at the lower level. If this is the case the efficient estimates of the quality differences in units of the differences at the lower level of n will be given by $\frac{1}{2}$ the differences of

$$n_1q_0 + 2n_2q_0, n_1q_1 + 2n_2q_1, n_1q_2 + 2n_2q_2$$

measured over all levels of p . Deviations from this supposed type of quality

effect, i.e. the interaction $Q.N$, will be given by the differences of

$$2n_1q_0 - n_2q_0, 2n_1q_1 - n_2q_1, 2n_1q_2 - n_2q_2$$

which are orthogonal to the above differences. (The reader will find it instructive to take some numerical example and check that the sums of squares for Q and $Q.N$, calculated from the above expressions, total to the sum of squares for the 3×2 table less the sum of squares for the $n_2 - n_1$ component of N .)

Similarly the interactions $Q.P$ and $Q.N.P$ will be given by the interactions of the two 3×3 tables containing the values of the above expressions for all levels of p .

If the experiment is arranged in blocks of nine plots, the ordinary type of $3 \times 3 \times 3$ confounding being employed, it will be found that both $Q.P$ and $Q.N.P$, if calculated as above, will be affected by block differences. The simplest procedure is to construct the standard 3×3 table for q and p , including the dummy treatments. The quantities in this table will be free from block effects, and consequently the 4 degrees of freedom for interactions will be compounded of $Q.P$, $Q.N.P$ and certain error components. They will therefore serve to test for interaction between q and p .

We can, however, improve on this procedure by constructing a 3×3 table of the quantities

$$[n_1p_0q_0] + [n_2p_0q_0] + \frac{1}{3} S_0(n_0)$$

etc., or better (if the quality effect is of the type considered above) of the quantities

$$[n_1p_0q_0] + 2[n_2p_0q_0] + \frac{1}{3} S_0(n_0) - \frac{1}{3} S_2(n_0),$$

$S_0(n_0)$ being the sum of the n_0 plots in blocks containing neither $n_1p_0q_0$ nor $n_2p_0q_0$, and $S_2(n_0)$ being the similar sum in blocks containing $n_2p_0q_0$. Both these sets of quantities are orthogonal to blocks and to the main effects and the other two-factor interactions, and there is little loss of information.

It might be thought that the three-factor interaction could be dealt with in the same way, but unfortunately the analogous expressions are not orthogonal to the above expressions for $Q.P$, owing to the n_0 terms. They will, however, form estimates of the three-factor interaction, though the tests of significance $Q.P$ and $N.Q.P$ will not be independent, and the error sum of squares cannot be deduced by subtraction.

The simplest way of obtaining an estimate for error is to include the three-factor interactions in the error sum of squares. If this is not considered advisable the analytical procedure appropriate to the ordinary $3 \times 3 \times 3$ design may be followed, utilizing dummy treatments and omitting the 6 additional degrees of freedom from error.

The above methods of procedure, though not exact, will suffice for most practical purposes. The reader who is interested in the general problem should consult (3) and (8), where exact methods are evolved for some examples of this type.

16. ARRANGEMENTS WITH SPLIT PLOTS.

16a. *Structure and analysis of split-plot designs.*

An experiment of any design may have its plots divided into two or more parts for subsidiary treatments. This procedure is of practical utility when treatments are included which are of such a nature that they necessitate large plots, as for example may occur in combined varietal and manurial trials, in which it is often inconvenient to use such small plots for the varieties as are practicable for the fertilizers.

The use of split-plots in randomized block experiments, however, results in a loss of information on the whole-plot treatments (with a compensating gain on the sub-plot treatments and their interactions with the whole-plot treatments), compared with the information which would be obtained in an ordinary factorial design using the same sub-plots, even without confounding, and the use of split-plot designs should therefore not be resorted to without good practical reasons unless the effects of the treatments to be associated with the whole plots are not of primary importance. On the other hand if the use of an ordinary factorial design would necessitate an arrangement in randomized blocks, whereas the use of split-plots enables a Latin-square design to be adopted for the whole-plot treatments, the latter design does not necessarily result in any loss of efficiency even on the whole-plot comparisons, owing to the generally higher efficiency of the Latin square.

The formal analogy between split-plot designs and ordinary confounded experiments will be immediately apparent. In split-plot designs main effects are confounded, instead of high-order interactions, the whole plots being analogous to the blocks of an ordinary confounded experiment. Analytically the important difference is that whereas in confounded experiments the small amount of information on the confounded interactions accruing from inter-block comparisons is ordinarily ignored, in split-plot experiments the information from whole-plot comparisons is retained, so that in all split-plot designs there are two different errors, one relating to the whole-plot comparisons and the other to the sub-plot comparisons.

The analysis of split-plot experiments is formally simple. The analysis of variance is divided into two parts. The first part is calculated from the yields of the whole plots, and furnishes errors and tests significance for the whole-plot treatments, exactly the same procedure being followed as in an ordinary randomized block or Latin square arrangement. The second part is calculated from the yields of the sub-plots, deducting those parts of the sums of squares which have already been accounted for in the analysis of the whole plots. This is equivalent to analysing the deviations of the sub-plots from their respective whole-plot means.

In order to make the mean squares of the two parts of the analysis comparable it is customary to work both parts *in units of a single sub-plot*. The sums of

squares of the first part (as calculated from the whole-plot totals) will therefore be divided by an additional factor equal to the number of sub-plots in a whole plot. In calculating the standard errors applicable to the total yields of whole plots the whole-plot error mean square must consequently be multiplied by this factor.

In the special case in which the whole plots are split into two parts only the differences between the pairs of sub-plots may be analysed directly in exactly the same manner as the totals of the pairs. The sums of squares from these differences will then also be divided by an extra 2. One extra degree of freedom representing the mean difference, i.e. the main effect of the treatment for which the split is made, and corresponding to the correction for the mean in the analysis of the totals, will be included in the analysis of the differences. The calculation of the total sum of squares of the experiment gives a check on the calculation of the totals and differences of the pairs and their sums of squares.

Many useful extensions of the split-plot type of design are available. In general, plots may be split into any number of units, and the resultant sub-plots may if desired be subjected to a further split, and so on indefinitely. Corresponding to each split a different estimate of error will appear in the analysis of variance.

The whole plots may be arranged in either randomized blocks or Latin squares. The treatments of the sub-plots will ordinarily be arranged at random within each whole plot. If confounding is resorted to it is not necessary to include all the sub-plot treatments in every whole plot. Designs of this type are exactly parallel to the more complex types of confounding already discussed, with main effects substituted for one or more of the confounded interactions.

Furthermore in certain cases it is possible to impose Latin-square restrictions on sets of sub-plots. Such designs are parallel to the designs already given under the name of quasi-Latin squares. By replacing interactions by main effects such squares are seen to yield a number of designs in which whole rows or both rows and columns are subjected to different treatments, most of the interactions of the Latin-square treatments with these being determined with full precision. Quasi-Latin squares which have both rows and columns subjected to different treatments may conveniently be called *plaid* squares, while if either rows or columns, but not both, are so treated they may be called *half-plaid* squares. The use of split-plot Latin squares in varietal trials is a further important application.

Examples of these extensions will be given at the end of the section. First, however, we will give an example of a simple split-plot design in randomized blocks.

16b. *Example : a varietal and manurial trial on oats.*

The results of this experiment have already been given in Section 9a. The plan and yields of the individual plots are given in Table 74, the analysis of variance in Table 75.

TABLE 74. VARIETAL AND MANURIAL TRIAL: PLAN AND YIELDS IN $\frac{1}{4}$ LB.

v_3	n_3	156	n_2	118	n_2	109	n_3	99	v_3
	n_1	140	n_0	105	n_0	63	n_1	70	
v_1	n_0	111	n_1	130	n_0	80	n_2	94	v_2
	n_3	174	n_2	157	n_3	126	n_1	82	
v_2	n_0	117	n_1	114	n_1	90	n_2	100	v_1
	n_2	161	n_3	141	n_3	116	n_0	62	
v_3	n_2	104	n_0	70	n_3	96	n_0	60	v_2
	n_1	89	n_3	117	n_2	89	n_1	102	
v_1	n_3	122	n_0	74	n_2	112	n_3	86	v_1
	n_1	89	n_2	81	n_0	68	n_1	64	
v_2	n_1	103	n_0	64	n_2	132	n_3	124	v_3
	n_2	132	n_3	133	n_1	129	n_0	89	
v_2	n_1	108	n_2	126	n_2	118	n_0	53	v_1
	n_3	149	n_0	70	n_3	113	n_1	74	
v_3	n_3	144	n_1	124	n_3	104	n_2	86	v_2
	n_2	121	n_0	96	n_0	89	n_1	82	
v_1	n_0	61	n_3	100	n_2	97	n_1	99	v_3
	n_1	91	n_2	97	n_2	119	n_3	121	

← Rows →

Area of each sub-plot: $\frac{1}{80}$ acre. (28.4 links \times 44 link rows.)

TABLE 75. VARIETAL AND MANURIAL TRIAL: ANALYSIS OF VARIANCE (SUB-PLOT BASIS).

			D.F.	Sum of squares	Mean square
Correction for mean			..	778336.06	
Whole plots	Blocks	..	5	15875.28	3175.06
	Varieties	..	2	1786.36	893.18
	Error	..	10	6013.30	601.33
Total			17	23674.94	
Sub-plots	Nitrogen	..	3	20020.50	6673.50
	N \times Varieties	..	2	321.75	53.63
	Error	..	45	7968.76	177.08
Total			71	51985.95	

The sums of squares for varieties, nitrogen, and their interactions are calculated from the two-way table (Table 34) in the manner explained in Section 9a. The sum of squares for blocks is calculated from the block totals in the ordinary manner, dividing by 12 after squaring, and the total sum of squares between whole plots is calculated from the whole-plot totals, dividing by 4 after squaring. The total sum of squares for the whole experiment is calculated directly from the yields of the 72 sub-plots. The whole-plot error is then obtained by subtraction of the sums of squares for blocks and varieties from the total sum of squares between whole plots, and the sub-plot error is obtained by subtraction of this total and the sums of squares for nitrogen and the interactions from the total sum of squares for the whole experiment. The formal analogy of this analysis with that of Table 12 should be noted.

It is immediately clear that the effect of nitrogen is definitely significant, but that the varietal differences do not approach significance. The deceptive appearance of the table of the yields of the treatment combinations (Table 76) in this respect should be noted. Here, although the differences between the varieties are not significant, the varieties fall in the same order, v_1, v_2, v_3 , at each level of n . This is characteristic of split-plot experiments in which the whole-plot error is substantially greater than the sub-plot error, being due to the fact that the same whole-plot errors affect all levels of the sub-plot treatments.

In the present example the interactions mean square is very decidedly below expectation, but not quite significantly so. Had it been significantly below expectation, this could of course only have been due to chance, unless there were some error or defect in the statistical analysis: for this reason if significantly sub-normal results occur repeatedly in any type of work the statistical procedure should be reviewed, both in its numerical and theoretical aspects.

16c. Calculation of standard errors.

Since there are two different errors applicable to whole-plot and sub-plot comparisons respectively, the calculation and use of the standard errors applicable to the yield totals of Table 34 require a little care. The varietal totals are totals of 6 whole-plots (= 24 sub-plots) and their standard error is therefore (from the whole-plot error mean square)

$$\sqrt{6 \times 4 \times 601.33} = \sqrt{24 \times 601.33} = 120.1$$

The nitrogen totals are totals of 18 sub-plots, and their standard error is therefore (from the sub-plot error mean square)

$$\sqrt{18 \times 177.08} = 56.4$$

The values in the body of the table are totals of 6 sub-plots, and in any comparison which involves the average effects of nitrogen and its interactions with varieties, but does not involve a mean varietal difference, the appropriate standard error of a single value is therefore

$$\sqrt{6 \times 177.08} = 32.6$$

Such comparisons include those between two values in the same line of the table or between the mean of two sets of values all in the same line, or any comparison made up of components of this type, and any interactions between varieties and nitrogen.

The conversion factor for the body of the table is $80/112 \times 4 \times 6$ and those for the margins are $\frac{1}{3}$ and $\frac{1}{4}$ of this. The final table of results is shown in Table 76.

Normally it will not be necessary to make comparisons between values in the body of the table which include any component of the mean varietal differences, and therefore in presenting the results it will usually be sufficient to give only the above three standard errors.

TABLE 76. MEAN YIELDS OF VARIETAL TRIAL IN CWT. PER ACRE.

	n_0	n_1	n_2	n_3	Mean
v_1	12.8	16.0	19.8	21.2	17.4
v_2	14.3	17.6	20.5	22.3	18.7
v_3	15.5	19.4	20.9	22.6	19.6
Mean	14.2	17.7	20.4	22.0	18.6
± 0.560					

S.E. of body of table (interactions and n effects only): ± 0.970 .

A comparison of this type may be required, however, when combining the results of experiments. We might, for instance, have a series of smaller trials on the same three varieties conducted at only two levels of nitrogenous manuring, 0 and 0.2 cwt. N per acre, and in the interests of uniformity we might then desire to abstract the mean of n_0 and n_1 from the results of the experiment under consideration. The standard error of these means can be derived as follows. Calculate the variance (the square of the standard error) of the mean of each pair of values from the standard error given in Table 76 for the body of the table. This is

$$\frac{1}{2} (0.970)^2 = 0.470$$

Also calculate the variance of the varietal means from this standard error, and subtract this from the actual variance of the varietal means given in the table. This gives

$$(0.894)^2 - \frac{1}{2} (0.970)^2 = 0.799 - 0.235 = 0.564$$

which is the additional component of error variance due to whole plots. Add these two variances together

$$0.470 + 0.564 = 1.034$$

and take the square root, 1.017, which is the required standard error. The point of this calculation is that the additional component of error due to whole plots is not increased by taking a mean over some instead of all the sub-plots in a whole plot.

16d. Efficiency.

It is immediately apparent that the whole plot comparisons are less precise than the sub-plot comparisons involving the same number of sub-plots, the ratio of the error variances being $601.33 : 177.08 = 3.40 : 1$. If instead of assigning varieties to whole plots we had completely randomized all 12 combinations of varieties and amount of nitrogen there would only be a single error. The expected value of this error can be found by the method of Section 7b, replacing each treatment mean square by the corresponding error mean square (Table 77).

This gives an error mean square of 254.22, so that the precision of the varietal comparisons would have been increased by complete randomization in the ratio $601.33 : 254.22 = 2.37$, while the precision of the nitrogen effects and its interactions with varieties would have been decreased in the ratio $177.08 : 254.22 = 0.70$.

TABLE 77. CALCULATION OF ERROR WITH COMPLETE RANDOMIZATION.

			D.F.	Sum of squares	Mean square
	Blocks		5	15875.28	
Remainder	Whole plots		12	7215.96	601.33
	Sub-plots		54	9562.32	177.08
	Total within blocks ..		66	16778.28	254.22

If the differences between varieties and the effects of nitrogen are of equal importance, then a completely random arrangement will clearly be the better, if not precluded by practical difficulties of sowing, etc. In certain cases, however, it may be that one set of main effects is of less importance than the other set and the interactions of the two sets. Thus, for example, the choice of variety might be dictated by other considerations than those of yield, in which case the primary function of the above experiment would be to determine the response to nitrogen and its possible variation from variety to variety. In this case the split-plot type of design is most appropriate. Similarly in an experiment including artificial fertilizers and dung there may be no particular point in determining with high precision the response to the dung (which is likely in any case to be of uncertain composition, and will certainly be applied in practice if available) though the variation in response to artificials in the presence and absence of dung may be of vital interest.

16e. *Confounding of interactions in split-plot designs.*

In addition to confounding the main effects of the whole-plot treatments, we may confound one or more interactions between the sub-plot factors with whole-plot differences, thus reducing the number of sub-plots in each whole-plot. The possibilities are very numerous, designs being most simply derived by applying different treatments to the blocks (now called whole plots) of ordinary designs. Thus in a combined varietal and manurial trial the varietal plots may be split into four for all combinations of the manurial factors n, p, k , the two sets of combinations (1), np, nk, pk and n, p, k, npk being assigned to different whole-plots, so that $N.P.K$ is confounded with whole-plots. With 6 varieties and 2 complete replications, each replication (12 whole-plots) being arranged in a block, the degrees of freedom in the analysis of variance will partition as in Table 78.

TABLE 78. DEGREES OF FREEDOM IN SPLIT-PLOT DESIGN.

Whole-plots				Sub-plots			
Blocks			1	N, P, K			3
Varieties			5	$N.P, N.K, P.K$..			11
$N.P.K$			1	$V \times$ manures ..			30
$V.N.P.K$			5	Error			36
Error			11				
Total			23	Total			72

We may, however, advantageously confound one of the degrees of freedom for $V.N.P.K$ with blocks, thus reducing each block to 6 whole-plots, one for each variety, and three for each of the two groups of manurial treatments. There will then be 3 degrees of freedom for blocks and 10 for whole-plot error. In similar designs with fewer varieties and whole-plots, in which the available degrees of freedom for whole-plot error are small, $N.P.K$ and $V.N.P.K$ may conveniently be included in the estimate of this error.

A further and most advantageous alternative is to arrange the whole-plots in a 6×6 Latin square. To do this, three complete replicates will be required. If one of the degrees of freedom for $V.N.P.K$ is confounded with rows it will be found that $N.P.K$ must be confounded with columns. Table 79 shows a square of this type after randomization, with numbers representing the varieties, and a dash the group of treatments (1), np , nk , pk .

TABLE 79. 6×6 LATIN SQUARE WITH SPLIT-PLOTS (6×2^3).

1	6'	4'	5'	2	3
4	3'	1'	2'	5	6
2	4'	5'	6'	3	1
3	5'	6'	4'	1	2
6	2'	3'	1'	4	5
5	1'	2'	3'	6	4

16f. Half-plaid Latin squares.

The treatment of whole rows or columns of a Latin square with a set of subsidiary treatments is a device which is very frequently useful. It is, however, only possible with certain special types of square analogous to the quasi-Latin squares already discussed.

At the outset it should be stressed that rows and columns must be completely randomized among themselves, as in quasi-Latin squares with confounded interactions. The arrangement of the replicates of the subsidiary treatments in blocks is therefore not permissible, but the additional degrees of freedom for error are a certain compensation for this disadvantage.

In order to ascertain if a square of the required type exists it is first necessary to see if there is a system of confounding which will give two suitable sets of degrees of freedom for confounding with rows and columns. If there is no confounding of interactions with the rows (these being subjected to the subsidiary treatments), i.e. if the number of treatment combinations of the remaining factors is equal to the side of the square, all that is required is an arrangement which confounds the whole factorial system (including subsidiary treatments) in randomized blocks of a size equal to the side of the square, i.e. an arrangement of the type that has already been enumerated for confounding in randomized blocks.

Thus, for example, in an 8×8 square with the rows sown with one or other of two varieties any one degree of freedom for the interaction of varieties with the other factors may be confounded with the columns. If the other factors form a $2 \times 2 \times 2$ system then the interaction chosen will naturally be $V.A.B.C$.

If four varieties are included the natural system of confounding with the columns will be of the type

$$V_1.A.B, V_2.A.C, V_3.B.C.$$

Partial confounding may be resorted to if desired, two sets of this type being confounded in a single square.

The actual construction of any required square can be easily effected. All that is necessary is to write down the sets of varietal and treatment combinations which confound the chosen interaction degrees of freedom, rearranging these sets so that the cross grouping in rows forms sets which each contain all combinations of the other treatments but only one variety.

Table 80 shows an 8×8 square for four varieties and a $2 \times 2 \times 2$ treatment system. The above set of interactions is confounded with the columns. (In order to exhibit the structure the rows and columns have not been randomized.) Such a square will not provide a very precise varietal test, but will furnish accurate information on possible interactions between the varieties and the other treatments.

TABLE 80. 8×8 HALF-PLAID SQUARE FOR FOUR VARIETIES.

v_1	1	8	3	6	4	5	2	7
v_1	8	1	6	3	5	4	7	2
v_2	3	6	8	1	7	2	4	5
v_2	6	3	1	8	2	7	5	4
v_3	4	5	7	2	8	1	6	3
v_3	5	4	2	7	1	8	3	6
v_4	2	7	4	5	3	6	8	1
v_4	7	2	5	4	6	3	1	8

Similar squares of other sizes are possible. Thus a 6×6 square may include two or three varieties in addition to the six treatment combinations forming a 3×2 system (factors a and b). If there are two varieties the arrangements of Section 13a will be required, partially confounding $V.B$ ($\frac{2}{3}$ information) and $V.A.B$ ($\frac{1}{3}$ information). If there are three varieties one of the arrangements of Section 13f will be required, or if two squares are available both arrangements may be used, giving $\frac{7}{8}$ information on $V.A$.

If there is confounding of interactions as well as subsidiary treatments with the rows, the construction of the squares requires a little more care. Thus, for instance, with a $3 \times 3 \times 3$ system of treatments and 3 subsidiary treatments applied to the rows one of the sets of confounded degrees of freedom shown in Table 43 would have to be adopted for the columns, and a set of the type

$$V, A.B.C, V.A.B.C,$$

for the rows.

TABLE 81. A 9×9 HALF-PLAID SQUARE.

<i>b</i>	<i>r</i> 8	<i>r</i> 3	<i>q</i> 1	<i>q</i> 5	<i>q</i> 9	<i>r</i> 4	<i>p</i> 7	<i>p</i> 2	<i>p</i> 6
<i>a</i>	<i>r</i> 6	<i>r</i> 7	<i>q</i> 8	<i>q</i> 3	<i>q</i> 4	■	<i>p</i> 5	<i>p</i> 9	<i>p</i> 1
<i>a</i>	<i>q</i> 2	<i>q</i> 6	<i>p</i> 4	<i>p</i> 8	<i>p</i> 3	<i>q</i> 7	<i>r</i> 1	<i>r</i> 5	<i>r</i> 9
<i>b</i>	<i>q</i> 4	<i>q</i> 8	<i>p</i> 9	<i>p</i> 1	<i>p</i> 5	<i>q</i> 3	<i>r</i> 6	<i>r</i> 7	<i>r</i> 2
<i>c</i>	<i>r</i> 1	<i>r</i> 5	<i>q</i> 6	<i>q</i> 7	<i>q</i> 2	<i>r</i> 9	<i>p</i> 3	<i>p</i> 4	<i>p</i> 8
<i>c</i>	<i>p</i> 5	<i>p</i> 9	<i>r</i> 7	<i>r</i> 2	<i>r</i> 6	<i>p</i> 1	<i>q</i> 4	<i>q</i> 8	<i>q</i> 3
<i>a</i>	<i>p</i> 7	<i>p</i> 2	<i>r</i> 3	<i>r</i> 4	<i>r</i> 8	<i>p</i> 6	<i>q</i> 9	<i>q</i> 1	<i>q</i> 5
<i>c</i>	<i>q</i> 9	<i>q</i> 1	<i>p</i> 2	<i>p</i> 6	<i>p</i> 7	<i>q</i> 5	<i>r</i> 8	<i>r</i> 3	<i>r</i> 4
<i>b</i>	<i>p</i> 3	<i>p</i> 4	<i>r</i> 5	<i>r</i> 9	<i>r</i> 1	<i>p</i> 8	<i>q</i> 2	<i>q</i> 6	<i>q</i> 7

Table 81 shows a square (randomized) of this type. This design has recently been proposed for a rotation experiment on sugar-cane, including 3 varieties (*p*, *q* and *r*), 3 quantities and 3 forms of nitrogenous fertilizer (combinations 1—9) and 3 levels of irrigation (*a*, *b* and *c*). It is intended that two squares should be laid down at each place, in different phases of the rotation, and that the experiment should be conducted at two or more places. The following sets of keys for the combinations 1—9 (Table 82), together with re-randomization, will serve to generate four squares confounding different sets of three-factor interactions.

TABLE 82. AMOUNT AND TYPE OF FERTILIZER.

Amount of fertilizer :		I			II			III			IV		
		0	1	2	0	1	2	0	1	2	0	1	2
Type of fertilizer	{ 1	1	4	7	1	7	4	1	3	2	1	2	3
	{ 2	2	5	8	3	9	6	4	6	5	7	8	9
	{ 3	3	6	9	2	8	5	7	9	8	4	5	6

With equal representation and no dummy treatments, half information would be obtained on the three-factor interaction of varieties, type and amount of fertilizers and three-quarters information on the other three-factor interactions. The existence of dummy treatments will modify these fractions somewhat.

The experiment originally suggested was one involving nitrogenous fertilizers only, but enquiry elicited (1) that the chief interest of the station was in varieties, (2) that irrigation was likely materially to affect the optimal level of manuring, and (3) that varieties and possibly the response to different forms of manuring, and (3) that varieties had already shown differences in their behaviour on good and poor soils and therefore might be expected to respond differently to manuring. It is quite probable, too, that varieties will behave differently under different conditions of irrigation. A factorial experiment is therefore essential if information of any real value is to be obtained. A half-plaid square is eminently suitable, since it would be exceedingly difficult to irrigate single plots differently.

As a further example the reader may construct an 8×8 square with a $2 \times 2 \times 2 \times 2$ system of treatments and two subsidiary treatments. He may also construct a set of 4×4 squares for four varieties, with four treatments (2×2) within the squares, sacrificing one-third the information on interactions between varieties and other treatments; and also a similar set of 4×4 squares for two varieties, retaining full information on all two-factor interactions.

16g. Plaid squares.

Instead of confining the confounding of main effects to rows only, different sets of main effects may be confounded with rows and with columns. Thus columns might be assigned to different varieties and rows to different cultivations. Upon randomization a typical Scotch plaid pattern will result.

Table 83 shows an example (before randomization) of this type of arrangement, comprising three varieties, three cultivations and a 3×3 system of treatments within the square. The following degrees of freedom are confounded:

Rows: $U, A.B.V (Y), A.B.U.V (4 \text{ d.f.})$

Columns: $V, A.B.U (X), A.B.U.V (4 \text{ d.f.})$,

the four-factor interactions being those derived from the interaction of the other confounded sets. The partition of the degrees of freedom will be that shown in Table 84. The remainder terms contain three- and four-factor interactions only.

TABLE 83. A 9×9 PLAID SQUARE.

	v_0			v_1			v_2		
v_0	1	6	8	9	2	4	5	7	3
	5	7	3	1	6	8	9	2	4
	9	2	4	5	7	3	1	6	8
v_1	8	1	6	4	9	2	3	5	7
	3	5	7	8	1	6	4	9	2
	4	9	2	3	5	7	8	1	6
v_2	6	8	1	2	4	9	7	3	5
	7	3	5	6	8	1	2	4	9
	2	4	9	7	3	5	6	8	1

TABLE 84. DEGREES OF FREEDOM IN THE 9×9 PLAID SQUARE.

Rows		Square	
U	2	A	2
Remainder	6	B	2
		Two-factor interactions.	24
Columns		Remainder	36
V	2		
Remainder	6	Total	80

As further examples of the plaid square the reader may construct the 8×8 square confounding:

Rows: $U, V.A.B, U.V.A.B,$

Columns: $V, U.A.B.C, U.V.A.B.C$

and a set of 4×4 squares for two varieties, two cultivations, and four treatments within the square. He may also convince himself that no simple 12×12 plaid square exists for two varieties, two cultivations, and a $3 \times 2 \times 2$ system of treatments within the square.

16h. Use of Latin squares with split plots in varietal trials.

In an ordinary varietal trial which does not include any other factors all comparisons are required with equal accuracy. When the varieties can be sown (or planted) in approximately square plots small numbers of varieties (up to

8 or so) can be conveniently arranged in Latin squares, while if the numbers are large (25 or over) the quasi-factorial designs described in the next section are suitable. In the intermediate range (10 to 24), Latin squares with split plots and *Graeco-Latin squares* (described below) provide a useful set of designs.

In a split-plot Latin square for 14 varieties, for example, the varieties are divided into 7 pairs, these pairs being arranged in a 7×7 Latin square, one of each pair being assigned at random to one half of each whole-plot. The analysis of variance will, as usual, be divided into two parts, the partition of the degrees of freedom being that shown in Table 85.

TABLE 85. 7×7 SPLIT-PLOT LATIN SQUARE: PARTITION OF DEGREES OF FREEDOM.

<i>Whole plots</i>			<i>Sub-plots</i>		
Rows	..	6	Varieties	..	7
Columns	..	6	Error (b)	..	42
Varieties	..	6			—
Error (a)	..	30	Total	..	49
		—			
Total	..	48			

There are two types of varietal comparison, one between varieties forming a pair, and the other between varieties not forming a pair. These have different errors, that of the former being calculated from the sub-plot error variance (*b*), and that of the latter from the mean of the two error variances (*a*) and (*b*). More generally, if each whole-plot is subdivided into *k* sub-plots, the error variance of any two varieties not occurring in the same set of *k* is given by the weighted mean of the variances (*a*) and (*b*), the weights being in the ratio 1 : *k* - 1.

16i. *The Graeco-Latin square.*

The main objection to the above type of design is that if the errors (*a*) and (*b*) are very unequal the comparisons between varieties in the same set and between varieties in different sets are by no means equal in accuracy. An alternative design, which overcomes this disadvantage at the expense of certain additional complication in the analysis, can be derived from a Graeco-Latin square.

A Graeco-Latin square consists of a pair of superimposed Latin squares, one formed of Latin, and the other of Greek letters, fulfilling the condition that every Latin letter occurs once and once only with every Greek letter, and vice versa. The two squares are thus mutually orthogonal, and a Graeco-Latin square is consequently derivable from any pair of squares of an orthogonal set. Graeco-Latin squares are known to exist for all numbers except even numbers which are not a multiple of 4. Of these latter numbers only 6 has been exhaustively investigated. For this number there is no such square.

If we take the Latin and Greek letters of a Graeco-Latin square to represent varieties (or other treatments) a design similar to that of a Latin square with split-plots results. The usual randomization process must be adopted, i.e. randomization of rows and columns and randomization of the Greek and Latin letter within each pair of plots. The letters should also be assigned to the varieties at random. Table 86 shows a 7×7 design after randomization.

TABLE 86. 7×7 GRAECO-LATIN SQUARE.

α f	β g	ζ d	e η	ϵ c	b δ	a γ
β e	f γ	c η	d a	ζ b	ϵ a	δ g
ϵ b	c ζ	g γ	a δ	β f	a e	d η
c δ	ϵ d	a β	γ b	a g	η f	ζ e
ζ a	b η	δ f	g e	γ c	d β	c a
d γ	e δ	b a	c β	η a	g ζ	f e
η g	a a	ϵ e	ζ f	d δ	γ c	β b

The analysis can be effected by forming two tables, one of the sums and one of the differences of the pairs of plots. These should be set out as in Table 87.

TABLE 87. ANALYSIS OF A GRAECO-LATIN SQUARE.

	Sums of pairs of plots						Differences of pairs of plots (Latin minus Greek)					
	a	b	c	d	Total	a	b	c	d	Total
α												
β												
γ												
....												
Total												

TABLE 88. 7×7 GRAECO-LATIN SQUARE: PARTITION OF DEGREES OF FREEDOM.

Table of sums				Table of differences			
Rows of square	6	Total (Latin-Greek)	..	1	
Columns of square	6	Latin letters	..	6	
Latin letters	6	Greek letters	..	6	
Greek letters	6	Error (b)	..	36	
Error (a)	24				
Total	48	Total	..	49	

The analysis of variance follows the lines indicated in Table 88. Sums of squares for the differences of the varieties represented by the Latin letters and those represented by the Greek letters appear in both parts of the analysis, and are derived from the marginal totals of the tables of sums and differences. The "interactions" of both tables give the estimates of error (a) and (b) between

"whole plots" and "sub-plots" respectively, corresponding to the errors (a) and (b) of Table 85. Thus estimates of the two types of error are separately obtained.

If the mean yields of the different varieties are taken as estimates of the varietal differences the error variance of the difference of two varieties in the same letter group (i.e. both Latin or both Greek) is, as before, derived from the mean of the two variances (a) and (b), while the error variance of the difference of two varieties in different letter groups is derived from a weighted mean of the two variances, the weights being in the ratio $p - 1 : p + 1$. The mean yields may be immediately obtained from the sum of the two sets of column totals, and the difference of the two sets of row totals, of Table 87.

It is worth noting that if the two error variances (a) and (b) are widely different more accurate estimates of the varietal differences may be obtained by taking a weighted mean of the estimates derived from the sum and difference tables of Table 87.

16j. The hyper-Graeco-Latin square.

Similar designs with the whole plots split into three or more parts may be constructed by the use of three or more squares from an orthogonal set. Such designs may be called *hyper-Graeco-Latin squares*.

The analysis of variance follows lines similar to that of a Graeco-Latin square, but the sums of squares cannot be derived from two-way tables. The

TABLE 89. ANALYSIS OF A HYPER-GRAECO-LATIN SQUARE.

	Latin letters			
Variety totals:	[a]	[b]	[c]
Whole plot totals:	[w _a]	[w _b]	[w _c]
	$k[a] - [w_a]$	$k[b] - [w_b]$	$k[c] - [w_c]$

simplest procedure is to set out the varietal totals for each group of letters (Latin, Greek, etc.) as in Table 89, and also the corresponding totals of the whole plots containing the varieties *a*, *b*, etc. (denoted by $[w_a]$, $[w_b]$, etc.). The difference of the second line from k times the first line is then taken. The second line (of the Latin letter table) provides estimates of the differences of the Latin letters derived from differences of whole plots, while the third line provides estimates derived from sub-plot differences. The sums of squares of the deviations, divided by pk and by $pk(k - 1)$ respectively, give the two sums of squares corresponding to the two sets of $p - 1$ degrees of freedom for the Latin letters in the whole-plot and sub-plot parts of the analysis respectively. The sums of squares for the Greek, etc., letters are derived similarly. The $k - 1$ degrees of freedom for the contrasts of the k groups of letters are derived from the contrasts of the total of the first line of Table 89 and the corresponding totals for the Greek, etc. letters.

The error variance of the difference of the mean yields of two varieties in the same group is derived from a weighted mean of the variances of whole and sub-plots, the weights being in the ratio $1 : k - 1$, and that of two varieties not in the same group is derived from a second weighted mean, the weights being in the ratio $p - 1 : p(k - 1) + 1$.

17. VARIETAL TRIALS—QUASI-FACTORIAL DESIGNS.

Plant breeders frequently wish to compare a large number of new strains—numbers such as 100 to 1000 are by no means uncommon. With such a large number of varieties arrangements in randomized blocks including all the varieties will usually be ineffective in eliminating fertility differences, while Latin squares are clearly impossible. The classical way of arranging such trials is by the use of "controls," i.e. plots growing a standard variety. These may be arranged either systematically or at random. Recently, however, new methods of arranging such trials have been devised, which make possible the use of blocks containing only a few plots, or, what is even more useful in many cases, the use of Latin squares. Most of these designs may be classified as "quasi-factorial,"* since their structure can be derived from confounded factorial designs. Such designs are always more efficient than designs involving controls, and will also be more efficient than designs in ordinary randomized blocks when there are any considerable inequalities of fertility.

It would take us too far afield to describe all these designs in detail. We shall therefore merely give an outline of the more useful types, without any attempt to describe the methods of computation. The reader who wishes to utilize the designs should refer to the original papers, (11), (12) and (13), where he will find a full description, together with numerical examples of the computations.

17a. *The lattice.*†

This is the simplest of the quasi-factorial designs in randomized blocks. If we have, say, 90 varieties, numbered 1—90, the rows and columns of the two-way table (Table 90):

TABLE 90. SETS FOR LATTICE DESIGN.

1	2	3	4	5	6	7	8	9	10
11	12	13	14	15	16	17	18	19	20
21	22	23	24	25	26	27	28	29	30
31	32	33	34	35	36	37	38	39	40
41	42	43	44	45	46	47	48	49	50
51	52	53	54	55	56	57	58	59	60
61	62	63	64	65	66	67	68	69	70
71	72	73	74	75	76	77	78	79	80
81	82	83	84	85	86	87	88	89	90

divide the varieties into two groups of sets containing 10 and 9 varieties each respectively. In a lattice design the varieties in each set are arranged in the field in randomized blocks, each group of sets being replicated equally. Thus, for example, with 6 replications, each group of sets will be replicated 3 times, there being 27 blocks of 10 plots each, of which three will contain varieties 1—10, and 30 blocks of 9 plots each, of which three will contain varieties 1, 11, 21, 31, 41, 51, 61, 71, 81.

*I have previously used the term "pseudo-factorial," but "quasi-factorial" seems preferable both descriptively and etymologically.

†The name is new.

The design is parallel to a factorial design, each variety being representable by a combination of two factors, one at q levels corresponding to rows, and the other at p levels corresponding to columns. In the replications of the first grouping the main effects of the first factor are confounded with blocks, in the replications of the second grouping the main effects of the second factor are confounded. The main effects of one or both factors will enter into the comparison of any pair of varieties, and therefore there is some loss of information on all such comparisons, comparisons between varieties which have a set in common being slightly more accurate than comparisons which have no set in common. This loss of information must be taken into account when assessing the efficiency of the design. The *efficiency factor** for a $p \times q$ lattice is

$$\frac{pq - 1}{pq + p + q - 3}$$

In the most useful case, when $p = q$, i.e. when the sets form the rows and columns of a square, it is

$$\frac{p + 1}{p + 3}$$

It may be noted that in any case q should not differ widely from p .

If p and q are small the efficiency factor becomes somewhat small. For 25 varieties, for example, it is $\frac{5}{8} = \frac{3}{4}$. This means that if there were no reduction in error variance per plot by reduction of block size from 25 to 5 plots, a lattice design would only give $\frac{3}{4}$ of the information that would be given by an ordinary arrangement in randomized blocks of 25 plots. Of course it rarely happens that there is no reduction in error variance, though the reduction is sometimes small. Moreover there is no reason why the information accruing from the block comparisons should not be taken into account, provided that the experiment has sufficient replications to give an adequate estimate of error for the inter-block as well as the intra-block comparisons. This procedure will recover most of the lost information and makes the design much more attractive for a moderate number of varieties.†

In order to utilize the information from inter-block comparisons, and to make these as accurate as possible, all the blocks forming a complete replication should themselves be arranged in a compact block on the ground. Pairs of these replications should contain one replication in each grouping, assignment of the grouping being at random within the pair. The sets should be assigned at random to the blocks of each replication.‡ Moreover the numbers of Table 90 (or the position within the table) should be assigned at random to the varieties.

*Defined as the ratio of the variance of a varietal comparison in a design in ordinary randomized blocks to the average variance in a lattice design occupying the same number of plots and having the same error variance per plot.

†This procedure is not discussed in the papers referred to above, but it is hoped to publish something on the matter shortly. In the simplest cases the additional computation required appears to be very small.

‡This method of arrangement is somewhat different from that of the example of (11), in which the use of inter-block comparisons was not envisaged.

17b. Triple and balanced lattices.

If the number of varieties is a perfect square, and a square lattice is constructed as above, it is always possible to superimpose a Latin square on this square. The letters of this Latin square may be used to denote a third group of sets, which may be arranged in randomized blocks in the same manner as the other two groups. We thus arrive at what may be called a triple lattice. It will be noted that all three groups of sets bear exactly the same orthogonal relationship to one another, every set of each group containing one and only one variety from every set of the other two groups.

The advantage of introducing a third grouping is that the efficiency factor is increased, being $\frac{p+1}{p+2\frac{1}{2}}$ instead of $\frac{p+1}{p+3}$.

If the number p is such that a full set of orthogonal Latin squares exists, further groupings corresponding to these squares may be made. When all the $p-1$ squares are used (giving $p+1$ groupings) complete balance is attained, comparisons between every pair of varieties being of equal precision. The efficiency factor of a balanced lattice is $\frac{p}{p+1}$. This corresponds to the fact that in each replication $p-1$ degrees of freedom out of the total of p^2-1 are confounded, so that the loss of information (blocks being completely ineffective) is

$$\frac{p-1}{p^2-1} = \frac{1}{p+1}.$$

This is a property of balanced arrangements, which has already been referred to.

Full sets of orthogonal squares are known to exist for all prime number and for $p=4, 8$ and 9 . No such set exists for $p=6$. For prime numbers the method of construction is very simple, each line of the first square being derived from the previous line by moving the letters one column to the right, each line of the second square by moving the letters two columns to the right, and so on. Sets of 8×8 and 9×9 squares are given in *The Design of Experiments* (2nd edition). The 10 groups for 81 varieties may also be derived by the successive transformation given in Section 14c of the square of Table 51. The first and second numbers of the treatment combinations and the rows and columns of each square give the 10 different groupings. The transformation given in Section 14c for the 8×8 square of Table 33 generates the groupings for 64 varieties in a similar manner, except that in the fourth square only the grouping given by the columns is required.

In all these lattice designs only a single replication of each grouping is necessary for the statistical reduction of the results, provided that information from inter-block comparisons is not required, but the actual number of replications will depend on the degree of precision desired, and will usually exceed these minimal requirements except in the case of balanced lattices.

17c. Lattice squares.

Instead of arranging the sets of a balanced lattice in randomized blocks, the groups of sets may be taken in pairs, and for each pair a square may be

constructed having its rows formed of the sets of one group and its columns of the sets of the other group. If p is odd, $\frac{1}{2}(p+1)$ squares will be required for balance, but if p is even each group must be included twice to give $p+1$ squares. If the rows and columns of each of these squares be rearranged amongst themselves in random order, and the resultant squares set out on the ground, we shall have an arrangement which is in essence a set of Latin squares with the quasi-factors confounded with rows and columns.

There is, of course, no absolute necessity for designs of this type to be balanced, but the attainment of balance, at any rate when p is odd, does not demand an excessive number of replications, and simplifies the computations and the interpretation of the results.

Table 91 shows a balanced set of three lattice squares for 25 varieties (before randomization of rows and columns).

TABLE 91. BALANCED SET OF LATTICE SQUARES FOR 25 VARIETIES.

Square I					Square II					Square III				
1	2	3	4	5	1	13	25	7	19	1	15	24	8	17
6	7	8	9	10	20	2	14	21	8	18	2	11	25	9
11	12	13	14	15	9	16	3	15	22	10	19	3	12	21
16	17	18	19	20	23	10	17	4	11	22	6	20	4	13
21	22	23	24	25	12	24	6	18	5	14	23	7	16	5

The method of construction of similar sets for other prime numbers should be apparent from a study of this table. Sets of squares for 64 and 81 varieties are provided by the transformation given in Section 14c of the squares of Tables 33 and 51, together with the square formed by arranging the varietal numbers in systematic order, as in the first square of Table 91.

These lattice squares are particularly attractive, since they enable the advantages of Latin square design to be utilized, whereas the comparisons within the sets of an ordinary lattice by means of Latin squares instead of randomized blocks would require more replications than are usually available. The efficiency factor is, however, somewhat low, being

$$\frac{p-1}{p+1},$$

as is easily verified from the property referred to above. With 25 varieties it has the value of $\frac{2}{3}$. The average increase in precision with 5×5 Latin squares in the Rothamsted experiments has been found to be $2.5 : 1$, so that the average net gain in precision on similar land by the use of lattice squares instead of ordinary randomized blocks for 25 varieties may be expected to be $1.67 : 1$ or 67 per cent. This average gain will be somewhat increased by utilizing inter-row and column comparisons in those experiments in which the land is found to be very uniform.

17d. Three-dimensional lattices.

Instead of arranging the varietal numbers in a two-way table, as in Table 21, they may be arranged in a three-way table, i.e. spatially in the form of a cube

or cuboid. A three-dimensional lattice, defining three groups of sets, may then be constructed by taking lines parallel to the edges of this cube or cuboid. Thus if there are $p \times q \times r$ varieties there will be pq sets of r varieties, pr sets of q varieties, and qr sets of p varieties. With $p = q = r$ there will be three groups of p^2 sets of p varieties. Thus an arrangement for $p \times q \times r$ varieties in blocks of p , q and r plots, or for p^3 varieties in blocks of p plots, is provided. The efficiency factor in the latter case is

$$\frac{2(p^2 + p + 1)}{2p^2 + 5p + 11}$$

Using a three-dimensional arrangement of p^3 varieties in the form of a cube, we may also obtain three groups of p sets of p^2 varieties by taking layers of this cube parallel to each of the faces in turn. The p^2 varieties of each set may be compared by means of a set of lattice squares, the use of two of the three groups being all that is really necessary. We thus arrive at an arrangement for p^3 varieties in $p \times p$ lattice squares. The efficiency factors are

$$\frac{p-1}{p+1} \cdot \frac{p^2+p+1}{p^2+p+3} \text{ and } \frac{p-1}{p+1} \cdot \frac{p^2+p+1}{p^2+p+2\frac{1}{2}}$$

respectively, according as two or three groupings in sets of p^2 are taken, the total number of replications required (p odd) being $(p+1)$ and $\frac{3}{2}(p+1)$ respectively.

17e. *Non-factorial designs : balanced incomplete blocks.**

In all the designs so far considered the number of treatment combinations is some multiple of the number of plots in a block or in a row or column of a Latin square, and moreover each replication of the design falls wholly in one set of blocks or rows or columns. There is a further useful family of designs in randomized blocks which does not in general fulfil these conditions. This is the family conforming to the condition that every pair of treatment combinations shall occur together in the same number of blocks. These designs are balanced, all treatment comparisons being of equal accuracy. Balanced lattices are members of this family, and other members are derivable from certain of the confounded designs already discussed. There are, however, many other members of the family which are not so derivable. The series of chief interest to the agronomist is a set of designs for $p^2 + p + 1$ varieties in blocks of $p + 1$ plots, with $p + 1$ replications. The structure of this set of designs is dependent on that of the orthogonal sets of $p \times p$ squares. They can be derived from the corresponding balanced lattices by adding one new variety to each block (the same variety being added to all the blocks of one grouping), and forming an additional block from all the $p + 1$ new varieties.

Balanced incomplete blocks are described in (5) and (12), and we shall not discuss them further here.

*Previously called symmetrical incomplete randomized blocks.

17f. The introduction of additional treatments in quasi-factorial designs.

The designs described in this section require a large number of blocks, and the possibilities of using these blocks as plots for additional treatments should not be lost sight of. If, for instance, there are six replicates of a simple lattice design, there will be sets of three blocks containing identical varieties, and these might be used as plots to compare three additional treatments and to ascertain whether the varieties interacted with these treatments. It will be noted that interactions between the additional treatments and the sets of varieties will inflate the inter-block error. This source of disturbance can be allowed for if necessary, but frequently it will not be sufficiently large to be of any moment.

NOTES

NOTE 1. NUMBER OF FIGURES REQUIRED IN THE COMPUTATIONS AND RESULTS.

It is a common fault in numerical work to retain too many figures both in the results and the intermediate calculations. On the other hand certain calculations require considerably greater accuracy than others, e.g. in the correction for the mean in the analysis of variance a large number of figures must be retained. There is not space here to give any detailed discussion of the matter, but the following hints may be of assistance.

(i) *Significant figures.*

The number of significant figures is the number of figures counting from the first figure not zero and excluding terminal zeros. Thus 237, 0.00237, 23700 all contain three significant figures.

(ii) *Observed yields, etc.*

Only three significant figures need be retained if the standard error of a single observation is not less than 3—5 per cent. of the mean (as in the yields of field plots). It pays to round off if the field results are given to greater accuracy. Fractions are best decimalized, as working in units of a quarter or a half of the ordinary units of measurement introduces dangerous possibilities of error. When a computing machine is used working means are best avoided, especially if they are such as to introduce negative numbers.

(iii) *Analysis of variance.*

Sufficient figures should be retained in the sums of squares to give four significant figures in the error sum of squares. In cases of doubt the retention of an extra figure or two does not seriously increase the work.

(iv) *Presentation of results.*

Three significant figures are normally sufficient in agricultural field experiments. In general the number of figures required depends on the accuracy of the final results.

(v) *Standard errors.*

A good 10 inch slide rule (three significant figures) will give all necessary accuracy, and is very convenient, since square roots may be read directly.

NOTE 2. NUMERICAL DIVISORS IN THE ANALYSIS OF VARIANCE, ETC.

The sum of squares corresponding to any single degree of freedom is obtained by squaring some quantity Q which is the sum of certain multiples (positive, negative and zero) of the plot yields. The divisor d by which Q^2 must be divided is equal to the sum of the squares of these multipliers. In the special but common case in which the multipliers are all +1, -1 or 0 the divisor is equal to the number of plot yields going to make up Q .

Technically Q is said to be a *linear function* of the plot yields y_1, y_2, \dots i.e.

$$Q = l_1 y_1 + l_2 y_2 + \dots$$

where l_1, l_2, \dots are numerical quantities (the above multipliers), so that

$$d = l_1^2 + l_2^2 + \dots$$

If more than one degree of freedom is involved there are several Q , and $\text{dev}^2 Q$ must be divided by a divisor d , which is calculated as above, *provided* no plot yield enters into more than one Q . If this occurs the difference of any two Q must be taken and a divisor calculated for this difference by the above rule. d is equal to one half of this divisor.

The estimates of the corresponding effects are obtained by dividing the Q by some divisor λd which depends on the conventions adopted. In the case of main effects and interactions of factors at two levels λ is equal to a half. With factors at more than two levels λ is equal to unity unless one or more of the interacting factors is at two levels only (see Section 13c for an example).

The error variance of Q is equal to d times the error variance of a single plot, and consequently the error variance of the estimate is $\frac{1}{\lambda^2 d}$ times the error variance of a single plot.

NOTE 3. ORTHOGONAL FUNCTIONS.

If the effects corresponding to two degrees of freedom are estimated from two quantities Q and Q' such that

$$\begin{aligned} Q &= l_1 y_1 + l_2 y_2 + \dots \\ Q' &= l'_1 y_1 + l'_2 y_2 + \dots \end{aligned}$$

as in Note 2, the two degrees of freedom are orthogonal if

$$l_1 l'_1 + l_2 l'_2 + \dots = 0$$

i.e. if the sum of the products of the corresponding multipliers of the plot yields is zero. With three degrees of freedom there are three such sums of products, which must all be zero, and so on.

Similarly two sets of degrees of freedom are orthogonal if the corresponding pairs of Q 's and Q 's are orthogonal, *provided* that no plot yield enters into more than one such pair.

NOTE 4. HINTS ON THE USE OF CALCULATING MACHINES.

(1) Arrange the computations so as to avoid having to write down intermediate steps: the transfer of numbers from the machine to paper, and back again to the machine, consumes a large amount of time, and introduces possibilities of error.

(2) Always compute sufficiently carefully to avoid mistakes. Checking should be regarded as an assurance that no errors exist, not as a method of correcting errors.

(3) In long computations, such as extensive sums of squares, record the value attained at suitable intervals, so as to facilitate the location of possible errors, but do *not* clear the machine.

(4) In calculating sums of squares or products accumulate the sum of the multipliers whenever possible, even if this sum is already known, either by means of a 1 on the right of the keyboard, or by means of the register provided on some machines for this purpose.

(5) Partial sums of the multipliers (such as block totals) may be obtained by recording the sum of the multipliers at the appropriate intervals, clearing this sum (but not the sum of squares) if convenient.

(6) In a sum of squares in which the sum is also being accumulated an occasional negative value (say - 123) may be treated by the process:

$$\begin{array}{r} 1\ 2\ 2\ 9\ 9\ 9\ 9\ 9\ 9 \\ 1\ 2\ 3 \\ \hline 1\ 5\ 1\ 2\ 8\ 9\ 9\ 9\ 8\ 7\ 7 \end{array}$$

the top line of figures being written on the keyboard. If there are a considerable number of negative numbers it is best to square all the positive numbers, record and clear their sum (but not the sum of squares), and then square all the negative numbers. Sums of products can be dealt with similarly.

(7) In covariance work with two variables the two sums of squares and *twice* the sum of products can be obtained simultaneously by the process:

$$\begin{array}{r}
 1230000456 \\
 1230000456 \\
 \hline
 01512901121760207936
 \end{array}$$

(A $10 \times 10 \times 20$ machine is required for three-figure numbers). If the sums of squares (together with the sums) are also calculated separately the sum of products will also be checked (but beware of negative numbers and errors of copying from the machine).

(8) In covariance work with more than two variables one sum of squares and one sum of products (or two sums of products) can be obtained simultaneously by writing two variables at opposite ends of the keyboard.

(9) In covariance work with more than two variables the most effective method of checking in many types of analysis is to construct an identical table of the sums (s) of the corresponding values of each variable. The various sums of squares of the s table provide a complete check, by reason of the identity

$$s^2 = (a + b + c)^2 = a^2 + b^2 + c^2 + 2ab + 2ac + 2bc.$$

More detailed checks are provided by the identities

$$as = a^2 + ab + ac$$

etc.

(10) If several divisions by the same divisor have to be performed it is best to multiply by the reciprocal of the divisor.

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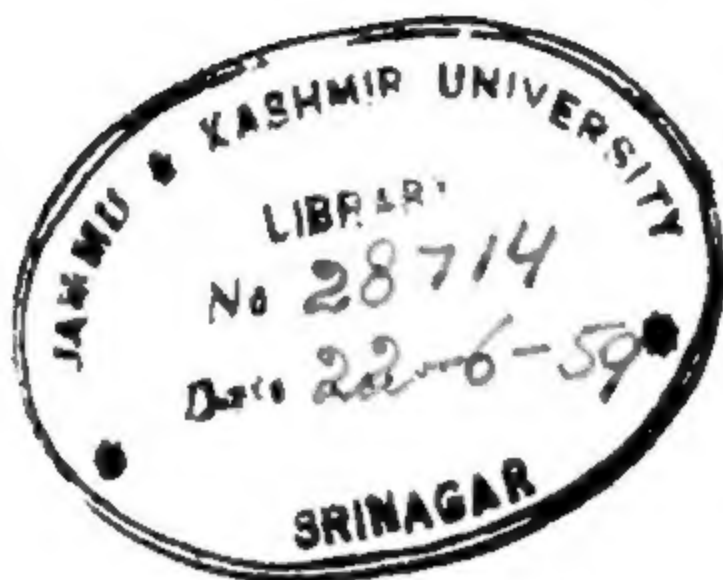
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